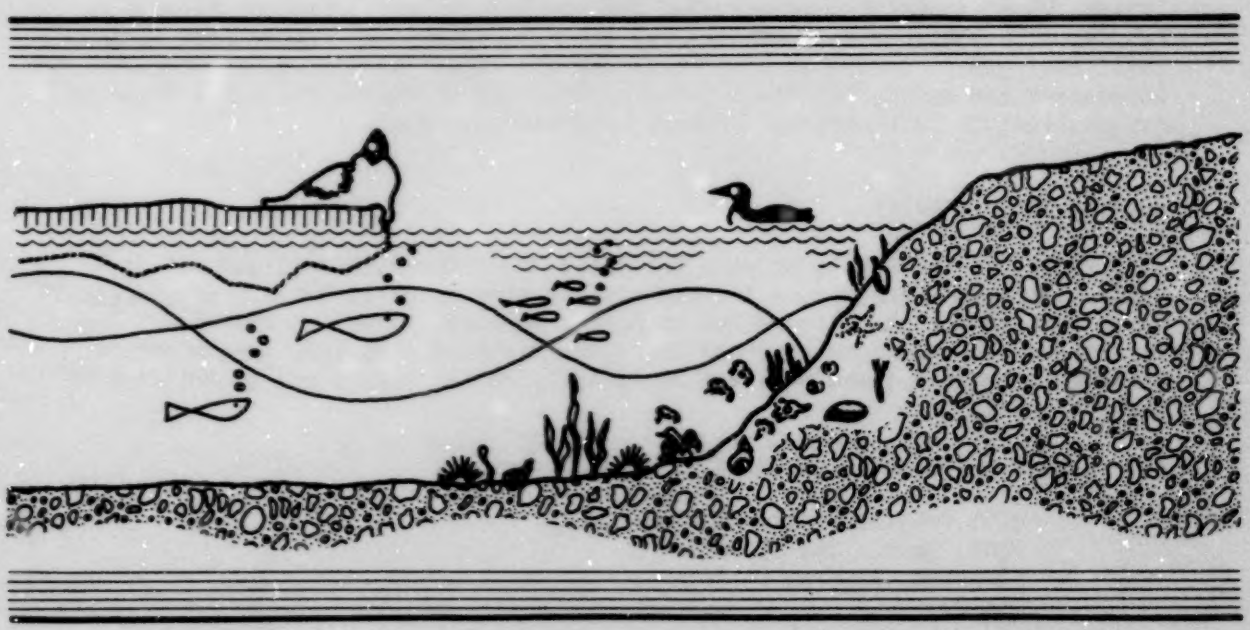
Environment Canada Environnement Canada

0058255E DEST PVM EXCH COPY 03
QH 91.8.04 W67
VOL ISS 81-5
WORKING REPORT SERIES (BAFFIN ISLAN
D OIL SPILL PROJECT (CANADA))

0058255E

MICROBIOLOGY

1. Effects of Oil on Bacterial Activity



Baffin Island Oil Spill Project

WORKING REPORT SERIES

365e

BIOS

CAPE HATT

1981 STUDY RESULTS

The Baffin Island Oil Spill Project

OBJECTIVES

The Baffin Island Oil Spill (BIOS) Project is a program of research into arctic marine oil spill countermeasures. It consists of two main experiments or studies. The first of these, referred to as the Nearshore Study, was designed to determine if the use of dispersants in the nearshore environment would decrease or increase the impact of spilled oil. The second of the two experiments in the BIOS Project is referred to as the Shoreline Study. It was designed to determine the relative effectiveness of shoreline cleanup countermeasures on arctic beaches.

The project was designed to be four years in length and commenced in 1980.

FUNDING

The BIOS Project is funded and supported by the Canadian Government (Environment Canada: Canadian Coast Guard; Indian and Northern Affairs; Energy, Mines & Resources; and Fisheries & Oceans), by the U.S. Government (Outer Continental Shelf Environmental Assessment Program and U.S. Coast Guard), by the Norwegian Government and by the Petroleum Industry (Canadian Offshore Oil Spill Research Association; BP International [London] and Petro-Canada).

WORKING REPORT SERIES

This report is the result of work performed under the Baffin Island Oil Spill Project. It is undergoing a limited distribution prior to Project completion in order to transfer the information to people working in related research. The report has not undergone rigorous technical review by the BIOS management or technical committees and does not necessarily reflect the views or policies of these groups.

For further information on the BIOS Project contact:

BIOS Project Office
#804, 9942 - 108 Street
Edmonton, Alberta
T5K 2J5

Phone: (403) 420-2592/94

Correct citation for this publication:

Bunch, J.N., Bédard, C. and Cartier, T., 1982, Microbiology: 1. Effects of Oil on Bacterial Activity - 1981 Study Results. (BIOS) Baffin Island Oil Spill Working Report 81-5: 82 p.

Baffin Island Oil Spill (BIOS) Project

Working Report 81-5

Microbiology

1. Effects of oil on bacterial activity - - 1981 study results

prepared for

Environmental Protection Service
Canadian Department of the Environment
Edmonton, Alberta

by

J. N. Bunch, C. Bédard and T. Cartier

March 1983

Arctic Biological Station
Department of Fisheries and Oceans
555 boul. St-Pierre
Ste-Anne-de-Bellevue, Québec
H9X 3R4

This report has also been issued as a Canadian Manuscript Report of Fisheries and Aquatic Science. The correct citation for this publication is:

Bunch, J. N., C. Bédard and T. Cartier. 1983. Abundance and activity of heterotrophic marine bacteria in selected bays at Cape Hatt, N.W.T.: Effects of oil spills, 1981. Can. MS Rep. Fish. Aquat. Sci. 1708: xiv + 82 p.

CONTENTS

	ABSTRACT	v
	RESUME	vii
	ACKNOWLEDGEMENTS	ix
	LIST OF FIGURES	xi
	LIST OF TABLES	xiii
1.0	INTRODUCTION	1
2.0	STUDY AREA	3
3.0	METHODS AND MATERIALS	5
	3.1 Sampling Procedures	5
	3.2 Dry Weight Determinations	5
	3.3 Chemical and Physical Oceanography	5
	3.4 Bacterial Counts	6
	3.5 Bacterial Activity	6
	3.5.1 Uptake of Glutamic Acid	6
	3.5.1.1 Theory	6
	3.5.2 Mineralization of Hexadecane and Phenanthrene	8
	3.6 Statistical Analyses	8
4.0	RESULTS	11
	4.1 The Water Column	11
	4.1.1 Total Counts of Bacterial Cells	11
	4.1.2 Counts of Colony-forming Units	12
	4.1.3 ^{14}C-Glutamic Acid Uptake	12
	4.1.4 Effect of Oil on ^{14}C-Glutamic Acid Uptake in the Water Column	13
	4.1.4.1 Surface Oil Release	13
	4.1.4.2 Dispersed Oil Release	14
	4.1.4.3 Effect of Petroleum and Corexit 9527 on Mineralization of ^{14}C-Glutamic Acid	15
	4.1.5 Most Probable Number of Oleoclastic Cells	15
	4.1.6 Hexadecane and Phenanthrene Mineralization	16

CONTENTS (continued)

	4.2	Sediments	17
		4.2.1 Total Counts of Bacterial Cells	18
		4.2.2 Counts of Colony-forming Units	18
		4.2.3 ^{14}C-Glutamic Acid Uptake	19
		4.2.4 Most Probable Number of Oleoclasts	20
		4.2.5 Mineralization of Hexadecane and Phenanthrene	21
5.0	DISCUSSION		23
6.0	CONCLUSIONS		29
7.0	REFERENCES		31
8.0	FIGURES		33
9.0	TABLES		49

ABSTRACT

Bunch, J. N., C. Bédard and T. Cartier. 1983. Abundance and activity of heterotrophic marine bacteria in selected bays at Cape Hatt, N.W.T.: Effects of oil spills, 1981. Can. MS Rep. Fish. Aquat. Sci. 1708: xiv + 82 p.

The effects of petroleum crude and a petroleum crude-dispersant mixture on the bacteria of the sediments and water column of nearshore locations at Cape Hatt, N.W.T., were assessed using data collected in 1981 before and after experimental oil releases and data acquired during a baseline study in 1980. In vitro experiments using petroleum crude, dispersant (Corexit 9527) or a mixture of both provided additional information concerning oil effects on bacteria. Despite some modifications, the collection and processing of samples were carried out in essentially the same way as in 1980.

During the 1981 field season, oleoclasts and oleoclastic activity were found to be widespread in both the water column and sediments. Oleoclastic numbers and activity did not appear to be affected by the oil releases. Water and sediment samples treated with Corexit 9527 in a concentration of 0.01% consistently showed lower levels of hexadecane mineralization than untreated samples. It is concluded that the addition of Corexit to spilled petroleum crude might severely limit biodegradation of the oil.

Analysis of variance was used to detect oil effects on total counts, counts of colony-forming units and V_{max}, turnover and (K+S) of glutamic acid uptake in the water column and sediments. Unless otherwise stated, an effect was judged significant when ($p<0.01$) was obtained. No change in either bacterial numbers or activity in the sediments can be definitely related to either of the oil releases. Following the dispersed oil release a transient and minor but significant decrease was seen in the V_{max} of the water column.

In vitro experiments showed that the respiration of glutamic acid was not significantly ($p>0.05$) affected by 0.1% by volume petroleum crude. Both 0.01% Corexit alone and a mixture of petroleum crude and Corexit caused significant decreases ($p<0.05$) in V_{max} and significant ($p<0.05$) increases in turnover and (K+S).

RESUME

Bunch, J. N., C. Bédard and T. Cartier. 1983. Abundance and activity of heterotrophic marine bacteria in selected bays at Cape Hatt, N.W.T.: Effects of oil spills, 1981. Can. MS Rep. Fish. Aquat. Sci. 1708: xiv + 82 p.

Les effets du pétrole brut et d'un mélange de pétrole brut et de dispersant, sur les bactéries benthiques et pélagiques, ont été évalués à des stations situées à proximité des côtes de Cape Hatt, T.N.O. Les données utilisées provenaient d'échantillons recueillis en 1981, avant et après des déversements expérimentaux de pétrole et d'échantillons accumulés au cours d'une étude préliminaire en 1980. Des renseignements additionnels concernant l'activité bactérienne en présence de pétrole brut, d'un dispersant (Corexit 9527) ou d'une combinaison des deux, ont été recueillis lors d'expériences in vitro.

Outre quelques modifications mineures, la collecte et le traitement des échantillons ont été réalisés essentiellement comme en 1980. Durant la saison de terrain 1981, une flore oléoclastique active était répandue à travers la colonne d'eau et les sédiments. Ni le nombre ni l'activité des oléoclastes n'ont semblé être effectés par les déversements de pétrole. Les échantillons d'eau et de sédiments traités au Corexit 9527 à une concentration de 0.01% ont affiché, sans exception, des taux de minéralisation d'hexadécane inférieurs à ceux des échantillons non-traités. Il a été conclu que l'addition de Corexit au pétrole brut lors d'un déversement pourrait en limiter la biodégradation de façon drastique.

La méthode de l'analyse de variance a été utilisée pour détecter les effets du pétrole sur les comptes totaux ("total counts") de bactéries, les comptes de colonies cultivées sur gélose ("colony-forming units") et le V_{max}, le turnover et le (K+S) de l'incorporation de l'acide glutamique dans la colonne d'eau et les sédiments. A moins d'avis contraire, un effet était jugé significatif lors de l'obtention d'un ($p<0.01$). Aucun changement, ni dans le nombre de bactéries ni dans leur activité, n'a pu être rattaché de façon définitive à l'un ou l'autre des déversements de pétrole. A la suite du déversement du mélange pétrole-dispersant, une diminution éphémère, légère mais significative a été enregistrée pour le V_{max} de la colonne d'eau.

Les expériences in vitro ont démontré que la respiration de l'acide glutamique n'était pas affectée de façon significative ($p>0.05$) par la présence de pétrole brut à une concentration de 0.1% par volume. Le

Corexit à 0.01% ainsi qu'un mélange pétrole brut et Corexit ont tous deux eu pour effet, d'une part, de diminuer de manière significative ($p<0.05$) le V_{max} et, d'autre part, d'augmenter de manière significative ($p<0.05$) le turnover et le (K+S).

ACKNOWLEDGEMENTS

We thank F. Dugré, R. Harland, J. Laliberté, L. Martin and P. Pannunzio for their analyses and discussions. F. Dugré, R. Harland, J. Laliberté and L. Ménard capably participated in field collections. M. Durand, R. Fortier and C. Trudeau contributed to the preparation of the report. We thank W. Cross, D. Thomson and others of LGL Ltd. for their diving collection. The help of K. MacGregor and the camp crew was greatly appreciated. Hydrocarbon data from ERCO and Seakem Oceanography Ltd. were appreciated. Figures 1 to 6 and Figure 8 were prepared by G. Ferrand. F. Aird prepared Figure 7 and Figures 10 to 14. The logistical and financial support of the BIOS project office is gratefully acknowledged.

LIST OF FIGURES

1. Microbiological stations occupied at Cape Hatt during 1981. 35

2. Total counts (TC) of bacterial cells determined in water samples collected at Cape Hatt, 1981. 36

3. Counts of colony-forming units (CFU) determined in water samples collected at Cape Hatt, 1981. 37

4. Maximum velocity (V_{max}) of glutamic acid uptake determined in water samples collected at Cape Hatt, 1981. 38

5. Turnover of glutamic acid uptake determined in water samples collected at Cape Hatt, 1981. 39

6. (K+S) of glutamic acid uptake determined in water samples collected at Cape Hatt, 1981. 40

7. Disintegrations per minute (dpm) obtained from water samples after 60 days incubation with ^{14}C-hexadecane. 41

8. Disintegrations per minute (dpm) obtained from water samples after 20, 40 and 60 days of incubation with ^{14}C-hexadecane. 42

9. Total counts (TC) of bacterial cells determined from sediment suspension samples collected at Cape Hatt, 1981. 43

10. Counts of colony-forming units (CFU) determined from sediment suspension samples collected at Cape Hatt, 1981. 44

11. Maximum velocity (V_{max}) of glutamic acid uptake determined from sediment suspension samples collected at Cape Hatt, 1981. 45

12. Turnover of glutamic acid uptake determined from sediment suspension samples collected at Cape Hatt, 1981. 46

13. (K+S) of glutamic acid uptake determined from sediment suspension samples collected at Cape Hatt, 1981. 47

14. Disintegrations per minute (dpm) obtained from sediment suspensions after 60 days incubation with ^{14}C-hexadecane. 48

LIST OF TABLES

1. Water column station occupations at Cape Hatt, 1981. 51

2. Sediment station occupations at Cape Hatt, 1981. 52

3. Determinations of total counts (TC) of bacterial cells from water samples collected at Cape Hatt during July 1981. 54

4. Determinations of colony-forming units (CFU), total counts (TC) of bacterial cells, maximum velocity (V_{max}), turnover (T) and (K+S) (see text) of glutamic acid uptake from water samples collected at Cape Hatt during August and September 1981. 57

5. Comparison of uptake parameters before and after the surface release of oil. 64

6. Hydrocarbon determinations from water samples collected at Cape Hatt on 27 and 28 August 1981. 65

7. Determinations of maximum velocity (V_{max}), turnover (T) and (K+S) (see text) of glutamic acid uptake from water samples collected at Cape Hatt on 27 and 28 August 1981. 66

8. Mineralization of glutamic acid in the presence of 0.1% by volume weathered Norman Wells (N.W.) petroleum crude with and without Corexit 9527 supplementation. 67

9. Most probable number (MPN) determinations of oleoclastic cells and maximum disintegrations per minute (dpm) obtained from water samples incubated with ^{14}C-hexadecane. 68

10. Comparison of disintegrations per minute (dpm) obtained from water and sediment suspension samples incubated with ^{14}C-hexadecane in the presence of different crudes. 72

11. Comparison of disintegrations per minute (dpm) obtained from water and sediment suspension samples incubated with ^{14}C-hexadecane or ^{14}C-phenanthrene. 73

12. Determinations of colony-forming units (CFU), total counts (TC) of bacterial cells, maximum velocity (V_{max}), turnover (T) and (K+S) (see text) of glutamic acid uptake from sediment suspension samples collected from surface sediment at Cape Hatt during August and September 1981. 74

13. Most probable number (MPN) determinations of oleoclastic cells and maximum disintegrations per minute (dpm) obtained from sediment suspension samples incubated with ^{14}C-hexadecane. 78

1.0 INTRODUCTION

The Baffin Island Oil Spill (BIOS) project at Cape Hatt, N.W.T. is a four year study designed to evaluate the effects of chemical dispersants during and after experimental oil releases in 1981. Integrated physical, chemical and biological studies during 1980 yielded baseline data. The reader is referred to the report of the Canadian microbiology studies (Bunch, Harland and Laliberté, 1981).

The objectives of the second year of study were to:

1. Repeat protocols of year one (1980) in the bays before and after releases to assess annual variations and variations due to oil releases.
2. Experimentally assess the use of radiolabelled phenanthrene and hexadecane as substrates for oleoclastic activity *in vitro*.
3. Experimentally assess the effects of dispersant, petroleum and petroleum dispersant mixtures on heterotrophic activity *in vitro*.
4. Relate microbiological data to hydrocarbon and physical and chemical oceanographic data and other biological data.

During the 1981 field season, experimental oil releases were conducted in two bays (see section 2.0); a surface release of 8% weathered Lago Medio petroleum crude on 19 August and a subsurface dispersed release of 8% weathered Lago Medio crude amended with Corexit 9527 on 27 August. Details of the releases can be found in Dickins (1982a).

In addition to *in vitro* experimental work during the field season, water samples were taken from the area of the dispersed oil release during and after the release to measure the immediate effects of dispersed oil on bacterial activities. This report summarizes data obtained during the 1981 field season and relates these data to those obtained in 1980.

2.0 STUDY AREA

During 1981, water and sediment samples were taken at eight stations at Cape Hatt (Fig. 1). Stations in bays 9, 10 and 11 were the same as those occupied in 1980 (Bunch et al, 1981). Because of concern about possible contamination of bay 10 (control bay) during the oil releases, bay 7 was chosen as an alternate control bay. On 3 August, a shore marker was placed by W. Cross of LGL Ltd. to indicate the centre of the bay; permanent markers were deployed at a later date to indicate stations 7 and 8 along a 12 m transect line.

The field season was divided into six numbered cycles for the water column and seven numbered cycles for the sediments representing the period from 3 August to 19 September (Tables 1 and 2). Cycles were defined as the time taken to sample all bays once. A spring sampling period from 17 July to 30 July included five cycles during which only samples for total counts of bacteria and environmental chemistry were collected. Whenever possible, collections were made simultaneously with those made by the Norwegian group of microbiologists. Sediment samples were divided for use by both groups.

Fifteen cubic metres of 8% weathered Lago Medio petroleum crude were pumped onto the surface of bay 11 on 19 August near high tide. The release area was contained by booms and prevailing winds drove the surface slick onto the beach as planned. Oil not beached was removed from the water surface by members of the Canadian Coast Guard. At low tide, the intertidal zone of the beach within the contained area was uniformly coated with oil from the release. Routine collections were made in bay 11 one day prior to the surface release and eight days afterwards. A supplementary collection was made on 21 August.

On 27 August, sixteen cubic metres of 8% weathered Lago Medio crude mixed with Corexit 9527 (10:1) were discharged from a dispersion pipe located perpendicularly to the shore and suspended one metre above the sediment at the south end of bay 9. The oil-dispersant mixture flowed north across bays 9 and 10, including the areas of stations 6, 5, 4, and 3, and out into Ragged Channel. Routine collections were made in bays 9

and 10 approximately one week before and two days after the dispersed release. Supplementary collections were made during the day of the release and the day after. The oil releases were executed by D. F. Dickins and details can be found in his report (1982a).

3.0 METHODS AND MATERIALS

Many of the procedures employed during the 1981 field season have been fully described in the 1980 Canadian microbiology report (Bunch, Harland and Laliberté, 1981). Following are changes of procedures used in 1980 and new procedures.

3.1 Sampling Procedures

Water column sampling procedures were the same as in 1980. Sediments were collected at all stations by LGL divers using modified 50 mL disposable syringes. For each station sampled, seven syringes on average were filled with surface sediments and capped for transportation back to the laboratory. Upon arrival, they were sorted and left to settle in a refrigerated area until processed. The top centimetre of all sediment cores taken from one station were combined and homogenized in a sterile Whirlpak bag (Fisher Scientific). With a 50 mL disposable syringe, a 20 mL wet subsample was measured and suspended in 2 L of filter-sterilized 10 m water. The 1.0% by volume (10^{-2}) sediment suspension was then manually agitated and the suspension maintained in a crushed ice bath on a magnetic stirrer while being processed. Remaining homogenized sediments were frozen and later shipped to Ste-Anne-de-Bellevue for total organic carbon and dry weight determinations.

3.2 Dry Weight Determinations

For each of the thawed sediment samples, three 1.0 mL subsamples were measured with a modified 3 mL disposable syringe, put into pre-weighed aluminum boats and dried at 60°C in an oven. After 48 h, the samples were transferred into a desiccator for a period of 30 min, weighed and results averaged.

3.3 Chemical and Physical Oceanography

Values for chlorophyll *a*, reactive phosphate and nitrate, ammonia and organic carbon in this report originated from Bunch, Bédard and

Cartier (1983). Materials for physical, chemical and microbiological analyses were collected simultaneously.

3.4 Bacterial Counts

In 1981, both plate count and total count data were obtained as in 1980. The technique for determining the most probable number of oleoclasts received some modifications. Forty mL samples of seawater or sediment suspension and tenfold serial dilutions up to 10^{-3} in 36 mL of filter-sterilized seawater were prepared in triplicate using serum bottles with rubber stoppers. Each bottle was supplemented with 0.8 mL of nitrate-phosphate solution, 40 µL of 22% weathered Lago Medio crude and 125 µL of n-(1-^{14}C)-hexadecane to yield a final activity of 6.25 µCi L^{-1}. The nitrate-phosphate concentrate yielded a final concentration of 1.0 g NH_4NO_3 and 0.1 g K_2HPO_4 per litre of sample water. Conditions of incubation were the same as in 1980. Incubations were terminated by the addition of 0.8 mL of 5.0 N H_2SO_4. Recovery and measurement of $^{14}CO_2$ evolved were carried out as in 1980.

3.5 Bacterial Activity

3.5.1 Uptake of Glutamic Acid

Bacterial heterotrophic potentials of L-[^{14}C(U)]-glutamic acid uptake were measured as in 1980. Because the results were expanded to include other kinetic parameters of glutamic acid uptake, the theory underlying the technique is presented below.

3.5.1.1 Theory

Kinetic parameters from the uptake of the glutamic acid substrate were calculated from a modified Michaelis-Menten equation (Dowd and Riggs, 1965):

$$\frac{D\mu t}{d} = \frac{(K+S)}{V_{max}} + \frac{A}{V_{max}}$$

where $D\mu$ = radioactivity added, d = radioactivity taken up, t = incubation time in hours, K = an uptake constant, S = concentration of the natural substrate, V_{max} = the maximum velocity of uptake, and A = concentration of the substrate added. Plotting $\frac{(D\mu T)}{d}$ against A yields a straight line where the reciprocal of the slope = V_{max}, y intercept = turnover time in hours (T), and x intercept = (K+S).

The maximum velocity (V_{max}), or potential of heterotrophic activity, is the velocity of uptake at which the substrate saturates the uptake system such that the velocity can no longer increase. V_{max} is an indication of the physiological state of the bacterial flora in that it demonstrates the potential ability of the flora to use a particular substrate, i.e. its degree of adaptedness to that substrate.

The value of (K+S) represents a combined value of the uptake constant and the concentration of the naturally occurring substrate in the water sampled. In a general way K may be considered as an affinity constant between cell and substrate. Specifically, it is the concentration of substrate required to drive the reaction at half-maximal velocity. A high value of (K+S) may suggest an unadapted population and/or a high concentration of natural substrate, while a very low value indicates an adapted population and/or a low value of natural substrate. Turnover (T) is the time required for the flora to deplete all the available natural substrate in a litre of the water sample. A very large value of T suggests a high concentration of natural substrate being consumed at a low velocity by an unadapted flora. A very low value of T suggests a highly adapted flora rapidly consuming a low concentration of natural substrate.

In addition to the above kinetic parameters, uptake and assimilation of a radioactive amino acid denotes conversion of dissolved organic carbon to particulate bacterial biomass (i.e. growth and multiplication). Measurement of released $^{14}CO_2$ provides an estimate of mineralization of an amino acid substrate to CO_2 and ammonia.

3.5.2 Mineralization of Hexadecane and Phenanthrene

Some modifications were included in the method for determining hexadecane mineralization. Forty mL aliquots of sample were used instead of 30 mL. Accordingly, added amounts of petroleum crude, nutrients and Corexit 9527 were increased to maintain the same concentrations as in the previous year. The final activity of n-(1-^{14}C)-hexadecane in the sample bottles was increased from 2.0 µCi L^{-1} to 6.25 µCi L^{-1}. No tris buffer was added.

In addition to those sets of bottles which received 22% weathered Lago Medio crude, separate sets received either weathered Norman Wells crude or weathered Lago Medio crude which was obtained from the group of Norwegian microbiologists. The Norwegian samples of crude were weathered by heating to 200°C. Water and sediment suspension samples for these sets were obtained from station 6. Sampling dates are specified in Table 10.

In yet another group of special sets, hexadecane was replaced with (9-^{14}C)-phenanthrene (Amersham Corp.). Final activity of phenanthrene in the sample bottles was the same as in n-(1-^{14}C)-hexadecane supplemented bottles (6.25 µCi L^{-1}). Samples for the phenanthrene mineralization study were obtained from station 6 during the August and September cycles.

3.6 Statistical Analyses

Kinetic parameters of glutamic acid uptake and their correlation coefficients were generated using computer programs developed by D. Burrage (Université du Québec à Montréal) and J. N. Bunch.

Spatial and temporal variations of the microbial parameters measured in the water column and sediments of Cape Hatt were studied with multiway analysis of variance (Sokal and Rohlf, 1969). All variates were transformed to common logarithms. Unless otherwise indicated, significance is expressed at the 1% level.

Data from the water column were grouped by bay, depth and cycle and three-way analysis of variance, including interactions carried out. If

significant bay-depth or cycle-depth interactions occurred, the analysis was repeated for each depth. Data from the sediments were grouped by bay and cycle, and two-way analysis of variance including interactions carried out. All possible two bay combinations were tested separately.

The source of significant bay-cycle interactions was carefully investigated since an oil release effect, if present, would be expected to yield such an interaction. When significant bay-cycle interactions occurred, summary graphs of the data (Figs. 2 to 6 and 9 to 13) were examined in order to detect which cycle(s) might be causing the interaction. Data from these cycles were removed from the data sets for analyses of variance and the analyses repeated.

Other analyses of variance carried out on various experimental data and data collected outside the normal sampling schedule are described in the results. A one-way analysis of variance was used to compare the results obtained at the two stations within a bay.

All analyses of variance were carried out using either the GLM or ANOVA procedures of the SAS computer package (Helwig and Council, 1979) available through the McGill University Computing Centre.

4.0 RESULTS

Cape Hatt ice conditions in 1981 were characterized by early break-up and late freeze-up. Field observations on 15 July indicated an already badly broken ice cover around Cape Hatt. By 17 July, when sampling started, ice cover varied between 2/10 and 5/10 in bays 9, 10 and 11, with ice moving back and forth with the tides. By 28 July, open water prevailed in Ragged Channel and in all bays sampled (Dickins, 1982b). Weather data were recorded twice daily during the whole 1981 season. These data, when considered together with field observations and temperature and salinity profiles, suggested that certain anomalies in the microbiological and chemical data were caused by storm surges. On 11 August, a storm with winds gusting to 33 knots considerably reduced operations at Cape Hatt. Temperature and salinity data recorded from water samples collected in bays 10 and 11 on 12 August underlined the presence of new water masses. These were characterized by a surface temperature about two degrees lower than that recorded on 10 August and salinity of 31°/$_{oo}$ as compared to a previous value of about 23°/$_{oo}$ (Bunch et al, 1983). Another storm on 4 September also altered the water masses sampled on 5 September, although to a lesser extent.

4.1 The Water Column

Statistical analysis of results indicates that no significant differences existed between depths at the same station, or stations in the same bay for most parameters. In most cases, values from the three depths sampled at both stations in a bay have been averaged and are presented in Figures 2 to 6. Data for all stations and depths are presented in Tables 3, 4, and 9.

4.1.1 Total Counts of Bacterial Cells

When sampling commenced in July, total counts (TC) of bacterial cells were high relative to values found in late August and September with a mean of $3.9 \times 10^8 \pm$ SE 0.4×10^8 cells L^{-1} recorded in the three

bays (Fig. 2). A maximum was reached in bay 11 with $6.0 \times 10^8 \pm$ SE 0.2×10^8 cells L^{-1} observed on 12 August. The high values in bays 10 and 11 on that date were probably due to a storm surge. Subsequently, numbers decreased steadily in all bays during the last half of August to a mean of $1.5 \times 10^8 \pm$ SE 0.1×10^8 cells L^{-1} recorded between 3 and 5 September. Samples taken during the last water cycle indicated a slight increase in all bays although cell numbers never reached levels observed earlier in the season when sampling started. No significant differences were seen between the bays when allowance was made for the storm surge on 12 August. It may be concluded that total counts of bacteria were unaffected by the two oil releases since no significant bay-cycle interactions were found.

4.1.2 Counts of Colony-forming Units

Counts of colony-forming units (CFU) were low in early August, the mean for all bays being $6.7 \times 10^5 \pm$ SE 1.0×10^5 colonies L^{-1} (Fig. 3). As summer progressed, counts increased only slightly. After 5 September, increases were noted in all bays, particularly bay 7. Values at that time ranged between $1.8 \times 10^6 \pm$ SE 0.4×10^6 and $6.0 \times 10^6 \pm$ SE 3.5×10^6 colonies L^{-1}. The two oil releases did not appear to affect counts of CFU. No significant differences between the bays were seen across the sampling season and no significant interaction was observed.

4.1.3 ^{14}C-Glutamic Acid Uptake

The maximum velocity (V_{max}) at which bacteria can potentially utilize glutamic acid is one of the kinetic parameters used to indicate the physiological state of heterotrophic floras. During the first sampling cycle, values of V_{max} for each of the bays were near their respective recorded maximum which was reached on 10 and 12 August for the four bays. At that time values ranged between 4.2 ± SE 0.4 and 6.5 ± SE 0.2 µg L^{-1} d^{-1} (Fig. 4). Following a subsequent drop in activity in all 4 bays, V_{max} values in the fourth cycle showed increases to levels similar to those observed earlier in August. As indicated in

Figure 4, September values were characterized by a slow decline with values equal to about half the seasonal maximum. With the first cycle of sampling (3-5 August) included in an analysis of variance, significant differences in V_{max} were seen in the bays. In the absence of the first cycle from the analysis, no differences or interaction were observed and V_{max} did not appear to be affected by the two oil releases. Turnover, a kinetic parameter, expressed as time (in days) taken by heterotrophs to mineralize the natural substrate (glutamic acid) contained in a litre of sampled water, remained relatively constant throughout the summer (Fig. 5). The means ranged between 1.4 ± SE 0.4 days in the first cycle to 2.3 ± SE 0.4 days in the third cycle, excluding cycle 5 where a large increase was recorded on 5 September. This increase was attributed to a storm on the previous day. Another kinetic parameter, (K+S) (see 3.5.1.1) did not vary greatly across the season, means ranging between 5.3 ± SE 0.5 and 7.4 ± SE 1.0 $\mu g\ L^{-1}$ (Fig. 6). Again, high values observed on 5 September were attributed to a storm.

4.1.4 Effect of oil on ^{14}C-Glutamic Acid Uptake in the Water Column

No effects of either oil release were noted in the seasonal data collected during the normal sampling cycles for uptake of glutamic acid. Observations were also made during and after the releases and from in vitro supplementation with petroleum crude.

4.1.4.1 Surface Oil Release

Water collections were made at stations 1 and 2 in bay 11 on 18 August as part of the second cycle of sampling. After the surface release of oil in bay 11 on 19 August, a supplementary collection of water samples from stations 1 and 2 was made on 21 August. The uptake of glutamic acid was determined in these samples and results compared to those obtained on 18 August. The comparison is seen in Table 5. On 21 August the mean of V_{max} determined from six depths decreased by one

fourth when compared to the same depths on 18 August, while turnover time (T) increased approximately threefold and (K+S) more than doubled. When the data from the second, third and fourth sampling cycles (including 18 August; see Figures 4, 5, and 6) from bays 7 and 11 were compared by a three-way analysis of variance, no significant interactions were observed for V_{max}, turnover time or (K+S). When the bay 11 data of 21 August were substituted for the data of 18 August in the same analysis, significant bay-cycle interactions were observed for turnover and (K+S) ($0.01>p>0.001$). Concentrations of high molecular weight hydrocarbons inshore from the microbiology stations in the area of the boom ranged from 0.007 to 0.73 mg L^{-1} in the upper 2 m of the water column between 20 and 22 August (Boehm, Fiest and Hirtzer, 1982). Negligible amounts of hydrocarbon were observed below 2 m although hydrocarbons were not determined in the area of the microbiology stations during this time. No significant difference in temperature, salinity or nutrients was observed during this time (Bunch et al, 1983).

4.1.4.2 Dispersed Oil Release

Water samples were collected from various depths in the water mass containing the dispersed oil on the day of the release and the day after. Concentrations of oil determined in the samples are seen in Table 6. Replicate samples were amended with ^{14}C-glutamic acid and incubated in the usual fashion (Bunch et al, 1981). The results are seen in Table 7 and can be compared to results from sampling both before and after the dispersed oil release in Table 4. In a three-way analysis of variance between bays 7 and 9 for cycles 3 (21 August), 4 (27-29 August) and 5 (10 September) no interactions were observed for the parameters of V_{max}, turnover or (K+S) (see Table 4). When data from 27 August (Table 7) were substituted for bay 9 data of 29 August, a significant bay-cycle interaction was determined for V_{max} ($0.01>p>0.001$), but not for turnover or (K+S), when the high value of 14.45 µg $L^{-1}d^{-1}$ for V_{max} from station 6 on 27 August was deleted from the

analysis. No interactions were observed when the data from 28 August were similarly treated.

4.1.4.3 Effect of Petroleum and Corexit 9527 on Mineralization of ^{14}C-Glutamic Acid

As part of the regular program across the sampling season, the mineralization of glutamic acid was measured in replicate sets of samples in the presence or absence of 0.1% Norman Wells crude, 0.01% Corexit 9527, or a mixture of both. In each instance, the kinetic parameters of V_{max}, turnover, and (K+S) (see 3.5.1.1) from the mineralization of glutamic acid to CO_2 were derived. The results are seen in Table 8. In a one-way analysis of variance, no significant differences in the three parameters were seen in the presence or absence of crude although changes were observed. In the presence or absence of Corexit 9527, significant differences were seen in V_{max} ($0.05>p>0.01$), turnover ($0.01>p>0.001$) and (K+S) ($0.05>p>0.01$). In the presence or absence of Norman Wells crude and Corexit, a significant difference was seen in V_{max} ($0.01>p>0.001$) and highly significant differences ($p<0.001$) were seen in turnover and (K+S). In the presence of crude and Corexit, turnover was significantly greater ($0.01>p>0.001$) than in the presence of crude alone as was (K+S) ($0.05>p>0.01$).

4.1.5 Most Probable Number of Oleoclastic Cells

The numbers of oleoclastic cells determined by the most probable number procedure (MPN) in water samples from 5 m, are presented in Table 9. An inadequate number of dilutions were employed in the MPN procedure in some cases. This affected the values for maximum numbers of oleoclasts obtained per litre from certain sampling dates, and underestimates only can be given for these dates. Variations of counts with time followed somewhat similar patterns in all four bays sampled. The sampling season started with low numbers between 0.525 and 27.5 x 10^3 cells L^{-1}. By the end of the second cycle, oleoclastic populations had increased to numbers exceeding 6.0 x 10^4 cells L^{-1} in all bays.

Prior to the dispersed oil release, counts were again low. From then on, and apparently independently from the dispersed oil release, numbers rose constantly until the last sampling in September when numerous underestimates of 6.0×10^4 cells L^{-1} were again recorded.

4.1.6 Hexadecane and Phenanthrene Mineralization

Results of hexadecane mineralization in water samples after several intervals of incubation are presented in Table 9 together with results from similar samples supplemented with Corexit 9527 and incubated for 60 days. Results of hexadecane mineralization are expressed as radioactive CO_2 (disintegrations per minute or dpm) evolved from labelled hexadecane. We did not consider it appropriate at this time to attempt a calculation of metabolized hexadecane. Table 10 presents a comparison of hexadecane mineralization in the presence of three different petroleum carriers, while comparisons of hexadecane and phenanthrene mineralization in replicate samples can be found in Table 11.

In the majority of samples, mineralization of hexadecane was not observed after 20 days of incubation. Most water samples showed the greatest mineralization after 60 days. A comparison of 60 day incubations is shown for the four bays in Figure 7. Hexadecane mineralization after a 60 day incubation period was near its recorded maximum early in August, with a mean value for all bays of $4.7 \times 10^5 \pm$ SE 1.9×10^5 dpm L^{-1}. This activity decreased from the end of the second cycle until the end of August to an average low of $0.9 \times 10^5 \pm$ SE 0.2×10^5 dpm L^{-1}. Subsequently, average dpm went up slightly until mid-September, at which time the counts shifted downward. The season ended with a mean recorded value of $2.4 \times 10^5 \pm$ SE 1.1×10^5 dpm L^{-1}. No pattern of activity emerged with respect to the oil releases although mineralization increased approximately threefold in bay 11 water samples from 12 September.

As seen in Table 9, the addition of Corexit 9527 generally depressed hexadecane mineralization by one or two orders of magnitude. Four examples are shown in Figure 8. In most cases, activity was

detected after a 40 day incubation in the absence of Corexit. Where Corexit was present, the lag period before detectable mineralization was extended and no appreciable activity was detected in the longest incubation of 60 days.

The use of weathered Norman Wells (N.W.) crude as a carrier for ^{14}C-hexadecane did not affect the lag period, but differences were noted in dpm per litre obtained after 60 days (Table 10). In most cases, counts were lower than those obtained using 22% weathered Lago Medio (L.M.) crude. When Norwegian Lago Medio (N.L.M.) crude was employed as a carrier for ^{14}C-hexadecane, lag periods similar to those seen with the other carrier crudes were observed. Norwegian Lago Medio was the designation given to a sample of weathered Lago Medio crude obtained from the Norwegian group of microbiologists. This crude had been weathered by bringing 8% weathered Lago Medio to a temperature of 200°C (K. Eimhjellen - personal communication). At this temperature, the crude should be considerably more than 22% weathered. In the three incubations maintained to 60 days, the mean result of hexadecane mineralization with the Norwegian Lago Medio carrier was approximately three times that obtained with the 22% weathered Lago Medio and twenty times higher than that obtained with weathered Norman Wells crude. A definitive comparison, however, would require a larger number of samples than that shown.

In the six water samples where phenanthrene mineralization was compared to hexadecane mineralization (Table 11), mineralization of phenanthrene was observed after 40 days in three cases only whereas hexadecane mineralization was observed in all cases. After 60 days, more than twice as much radioactivity was evolved from the hexadecane substrate than from the phenanthrene substrate.

4.2 Sediments

Results for total counts (TC), colony-forming units (CFU), V_{max} of glutamic acid uptake, turnover, (K+S), oleoclastic activity and oleoclast numbers obtained in the sediment of the four bays sampled for

microbiology during the months of August and September are presented in Figures 9 to 14 and Tables 12 and 13. Spatial and temporal variations of the above parameters, excluding oleoclastic numbers and activity, were examined using analysis of variance. Possible oil release effects were investigated by seeking the cause of significant bay-cycle interactions between control and release bays as outlined in section 3.6.

No significant differences between stations within bays were found in any of the parameters studied by analysis of variance. Therefore, on the basis of two sampling locations, it appears that within a bay at approximately 10 m depth, the sediments were relatively homogeneous at least with respect to the parameters measured.

4.2.1 Total Counts of Bacterial Cells

Significant differences were found between total counts obtained in bays 7 and 9 and bays 7 and 10 (Fig. 9). With a mean seasonal value of $1.5 \times 10^9 \pm$ SE 0.2×10^9 cells g^{-1} dry weight, twice the value found in bay 9 and 1.5 times that found in bay 10, bay 7 had the highest mean seasonal count of all bays sampled.

We detected no significant temporal variation in total counts in any of the bays. Since no significant bay-cycle interactions were found in any of our comparisons, it was concluded that the oil releases had no statistically detectable effects on total counts.

4.2.2 Counts of Colony-forming Units

The mean seasonal value of CFU in bay 7, $7.1 \times 10^6 \pm$ SE 1.3×10^6 colonies g^{-1} dry weight, was higher by an average factor of 5.6 than the mean CFU of all other bays. The bay 7-bay 9 comparison yielded a significant bay-cycle interaction. By repeating the analysis of variance without the data from cycle 2, during which time CFU levels in bay 7 reached their highest peak of the season, the interaction was eliminated. At the same time the existence of a significant difference between bays 7 and 9 was confirmed. The mean seasonal CFU value in bay

9 was significantly higher than that in bay 11 although the difference between these two bays was much less marked than the differences between bay 7 and all other bays (see Fig. 10).

Significant differences between cycles were found in bays 9 and 11. Except in the comparison between bays 7 and 9 mentioned above, no significant bay-cycle interactions were obtained, indicating that at most times the bays exhibited roughly parallel trends. For reasons that are not clear, bay 7 exhibited a striking rise in CFU from cycle 1 to cycle 2 which was in no way matched by similar changes in the other bays. We found no indications of any oil release effect on CFU.

4.2.3 ^{14}C-Glutamic Acid Uptake

V_{max} and turnover values for bay 7 were found to be significantly different from values obtained for these parameters in all other bays (Fig. 11, 12). The comparison of turnover in bays 7 and 9 yielded significant interaction betweeen bays and cycles. Repeating the analysis of variance following the removal of the data from cycle 4 confirmed the significant difference between bays 7 and 9 while eliminating the interaction between bays and cycles.

The mean seasonal value of V_{max} in bay 7, 37.72 ± SE 4.52 μg g^{-1} dry weight day^{-1}, was three times higher than the mean value in bay 9, 2.7 times higher than in bay 10 and 1.4 times higher than in bay 11. The mean seasonal value of turnover in bay 7, 0.98 ± SE 0.10 days, was half that in bay 9, 1.7 times lower than in bay 10 and 1.3 times lower than in bay 11. Bay 11 had significantly higher values of V_{max} than both bays 9 and 10. The comparison between V_{max} in bays 10 and 11 yielded a significant bay-cycle interaction. This interaction was no longer significant following the removal of the values for the first cycle from the data set for analysis of variance. Over the last five cycles the V_{max} in bay 11 was found to be significantly higher than that in bay 10.

Bays with a higher mean seasonal V_{max} and lower mean seasonal turnover values were characterized by higher mean seasonal total

counts. This, and the fact that (K+S) (Fig. 13) values did not vary significantly between bays, suggests that increases in glutamic acid uptake may be due, at least in part, to increases in bacterial numbers. This tended to be confirmed by highly significant ($p<0.001$) Spearman rank correlations between TC and V_{max} ($r = 0.678$) and TC and turnover ($r = -0.584$).

Significant differences between cycles in mean V_{max} were found in bays 10 and 11 and in mean turnover in bay 7. No significant temporal fluctuations were noted in (K+S). On the whole, the kinetic parameters related to glutamic acid uptake as measured in each bay tended to follow parallel trends across the season.

The only statistical evidence for a possible effect on the microbial flora of the dispersed oil release is given by the significant interaction obtained in the comparison of turnover values in bays 7 and 9. From cycle 3 to cycle 5 turnover in bay 9 decreased continuously. In bay 7, on the other hand, turnover increased between cycles 3 and 4 and then decreased. Bays 10 and 11 followed trends similar to bay 9. The sediments of bays 9, 10 and 11 were all contaminated by released oil (Boehm, Fiest and Hertzer, 1982). The petroleum crude may have caused a slight, short term, increase in glutamic acid uptake in bay 9 and possibly in bays 10 and 11. The possibility of a petroleum crude effect on glutamic acid uptake will be examined in greater detail in the discussion section.

4.2.4 Most Probable Number of Oleoclasts

Counts of oleoclasts obtained at the different stations across the season are given in Table 13. Oleoclasts were present in all sediment samples obtained for determination of oleoclastic activity and numbers. The lowest number of oleoclasts, 1.0 cell g^{-1} dry weight of sediment was obtained at station 4 on 7 August. An upper limit to the range of oleoclast number cannot be given since in 25 of 58 samples, the MPN samples containing the greatest dilution gave positive results. In these cases, only a lower limit for the most probable number of

oleoclasts can be given. It appears that in some cases the number of oleoclasts per gram dry weight of sediment exceeded 10^4. The lack of more precise estimates of oleoclast numbers precluded inter-bay and inter-cycle comparisons.

4.2.5 Mineralization of Hexadecane and Phenanthrene

In 53 of 58 sediment samples, n-(1-^{14}C)-hexadecane mineralization was observed after 20 days, the shortest period of incubation used in this study (Table 13). After 40 days all but one sample exhibited evolution of $^{14}CO_2$ from labelled hexadecane. The one exception, station 4 on 7 August, showed no activity after 60 days. The oleoclast count in this sample, 1.0 cell g^{-1} dry weight of sediment, was the lowest of all oleoclast counts obtained in sediments. The uniqueness of both the hexadecane mineralization value and the oleoclast count suggest that experimental error may have occurred. In any case, the general conclusion can only be that at Cape Hatt, oleoclastic activity is a widespread characteristic of the sediments.

Patterns of fluctuations in hexadecane mineralization in the different bays sampled after 60 days of incubation are summarized in Figure 14. We detected no obvious influence by the oil releases.

Corexit appears to have had a considerable effect on hexadecane mineralization. In 53 out of 58 samples supplemented with Corexit 9527 (Table 13) the amount of hexadecane mineralization after 60 days was considerably lower than in unsupplemented samples.

In sediment samples collected from station 6 on seven different dates, hexadecane was found to be degraded more rapidly than phenanthrene. In fact, after 60 days 4 out of 7 samples treated with ^{14}C-phenanthrene showed no $^{14}CO_2$ activity while all samples treated with ^{14}C-hexadecane showed such activity (Table 11).

Using the sediment samples described in the previous paragraph, we investigated the effect of different carrier petroleum crudes on the rate of hexadecane mineralization. The results are presented in Table 10. Taking into account the considerable variation in dpm values

obtained, there appeared to be no obvious difference in hexadecane mineralization when either Norman Wells or 22% Lago Medio crude was used as a carrier. Too few results from samples incubated with the Norwegian Lago Medio are available to effectively evaluate the effect of this crude on hexadecane mineralization in sediment samples.

5.0 DISCUSSION

The seasonal abundance and activity of bacteria in the bays at Cape Hatt were determined by a number of open water measurements in 1980 and 1981. Sampling intervals in 1981 were arranged to ensure that the oil releases occurred between sampling intervals.

In the water columns of the bays, the biological events of phytoplankton and bacteria proceeded in 1981 as in 1980 although ice break-up was somewhat earlier in 1981, and events were consequently earlier. There was no indication that levels of chlorophyll *a*, carbon or nutrients were significantly different in 1981 than in 1980 (Bunch *et al*, 1983).

That no effects of the oil releases were noted in the seasonal data from the water column was not unexpected since the waters of the bays were rapidly flushed of oil after both releases. Although significant interactions were found in the kinetic parameters of turnover and (K+S) of glutamic acid uptake in a supplementary sampling two days after the surface release of 8% weathered Lago Medio crude, these changes cannot be ascribed with certainty to the surface oil release. Oil concentrations in bay 11 during that time did not exceed 0.73 mg L^{-1} in the upper 2 m of the water column and were negligible below that depth. In a laboratory experiment (Table 8), glutamic acid mineralization was not significantly different in the presence or absence of 0.1% by volume weathered crude. Similar results were obtained by Alexander and Schwarz (1980) with 0.1% fresh crude. Contrary to the results of Alexander and Schwarz, Hodson, Azam and Lee (1977) found 15% inhibition of glucose assimilation in the presence of 0.8 mg L^{-1} of aqueous extracts of petroleum crudes, i.e. that fraction of fresh petroleum which is usually considered toxic. Griffiths, McNamara, Caldwell and Morita (1981) found that glucose and glutamic acid uptake were reduced significantly in water samples amended with 0.1% by volume fresh crude. They suggested that their study approximated an actual spill where similar concentrations of water-soluble (i.e. toxic) components of the oil would be in the water column directly adjacent to the spill. Weathering in a "real spill" situation was not discussed.

In the BIOS field study, Lago Medio crude weathered 8% was assumed to be representative of the degree of weathering which would be found in a recent accidental spill. A large proportion of the light fractions were still present. In this scenario, a decision could be taken to disperse the weathered surface slick.

During the dispersed oil release, oil concentrations ranged from about 200 mg L^{-1} at the dispersion pipe to an average of about 4.7 mg L^{-1} away from the pipe where water samples were collected for microbiological analyses (Table 6). During this time, with the exception of one high value, V_{max} of glutamic acid uptake decreased with respect to the control bay and a significant bay-cycle interaction was observed. This was not observed the following day when concentrations of oil had dropped to an average of 0.8 mg L^{-1}. Considerable adverse effects were seen in the benthic community (Cross and Thomson, 1982). Perturbation of bacterial uptake of glutamic acid in the water column, however, was short term. This effect could be attributed to the light fraction of the dispersed Lago Medio, but the effect of Corexit cannot be discounted.

The kinetic parameters of glutamic acid uptake were not significantly affected in in vitro experiments by supplementation with 0.1% by volume weathered crude (Table 8), although slight changes in these parameters were observed. The concentration of oil in water in these experiments was probably less than concentrations observed during the dispersed oil release away from the dispersion pipe. The experimental oil, moreover, was considerably more weathered than the oil used in the dispersed release. Replicate samples were significantly affected by the presence of 0.01% Corexit 9527 as were samples supplemented with an oil-dispersant mixture. No significant difference was seen between treatments of oil-dispersant or dispersant alone. V_{max} decreased approximately twofold and turnover and (K+S) increased by tenfold and twofold respectively over the control in samples supplemented with Corexit alone. Had V_{max} remained unchanged, this could be interpreted as a competitive inhibition of the uptake of the

glutamic acid substrate by a component of the dispersant. This unknown component would necessarily be taken up by cells through the same mechanism which transports glutamic acid across the cell membrane. Non-competitive inhibition or the loss of the transport mechanism is not suggested since (K+S) increased significantly over the control. Other than these usual explanations of alteration to the transport mechanism across bacterial membranes, the reason for the changes in all three parameters remains unclear. The presumed multiplicity of components in Corexit 9527 does not facilitate an easy interpretation of the data. Further, the concentration of Corexit in our experimental uptake studies was about three orders of magnitude greater than that observed during the dispersed release (assumed to be one-tenth of the concentration of the oil or approximately 0.5 mg L^{-1} on 27 August). An alteration of hexadecane mineralization by Corexit will be discussed below.

Emphasis has been placed on microbiological and chemical studies of subtidal sediments at Cape Hatt, since oil in the sediments can be expected to have a considerable residence time, particularly in bay 11 where continual subtidal oiling from the beached oil is anticipated.

Significant temporal and spatial variations were observed in measured microbiological parameters of the sediments. Bay 7, in particular, tended to differ, sometimes noticeably (e.g. mean CFU during the second cycle), from the other bays. In terms of both numbers of bacteria measured as colony-forming units or total counts and heterotrophic activity measured as V_{max} or turnover time of glutamic acid uptake, bay 7 exhibited the highest level of microbiological activity on a seasonal basis. Bay 7 tended to exhibit more spatial and temporal variations than the other bays. These facts combined to make bay 7 less than an ideal choice as a control bay. Therefore there is a possibility that the significant bay-cycle interaction observed in the analysis of variance of seasonal data from bays 7 and 9 was due not to an oil release effect but to differences in environmental conditions prevailing in the bays around the time of the dispersed oil release (see

below). On the whole, however, the microbiological characteristics of all bays tended to follow parallel courses through time.

Differences between bays and over time may be in part related to changes in the sedimentation rate of organic material from the water column. Parsons, Takahashi and Hargrave, (1977) have emphasized the importance of this material to the metabolism of benthic communities. Microorganisms are responsible for a large proportion of this metabolism. Furthermore, in a shallow marine environment such as the bays at Cape Hatt, resuspension of sedimented material might also be expected to affect benthic bacterial metabolism.

The increase in turnover observed in the sediments of bay 7 from cycle 3 to cycle 4 was not matched in any of the other bays. In particular, turnover in the sediments of bay 9 decreased sufficiently to cause a significant bay-cycle interaction to occur in the two-way analysis of variance of the data from bays 7 and 9. It is conceivable that the decrease in bay 9 was due to stimulation of glutamic acid uptake by the dispersed oil. Oil concentrations in bay 9 sediments reached approximately 0.01 mg g^{-1} (Boehm et al, 1982). Oil in the sediments may have acted in such a way as to decrease the value of K which could have led to the decreases in (K+S) and turnover seen between cycle 3 and 4 in bay 9. In this scenario the V_{max} of the bacterial population would not change, and in fact this was the case.

Although some evidence for stimulation by crude oil of the uptake of small organic molecules by marine sediment bacteria has been reported (Alexander and Schwarz, 1980; Griffiths et al, 1981), such stimulation has never been unequivocably demonstrated. Therefore, for our own data, we hesitate to endorse an explanation on the highly hypothetical grounds of stimulation by crude oil. Other hypotheses can be suggested to explain the interaction. For example, a decrease in glutamic acid concentration in the sediments of bay 9 but not in bay 7 is consistent with the changes seen in the kinetic parameters in these two bays between cycles 3 and 4. It should be noted that the magnitude of the decrease in turnover in bay 9 from cycle 3 to 4 was not greater than

decreases occurring at other times in the same bay or in other bays. It should also be mentioned that whatever the cause of the interaction, the effect was short-lived, in the order of a few days.

Numbers of oleoclastic cells and their activity in the water column and sediments did not appear to be affected by the oil releases. Nor was this unexpected. Hodson et al (1977) did not observe any enhancement of hydrocarbon oxidation or bacterial density 30 days after water enclosures had been amended with petroleum. Changes in sediment populations might be expected in the 1982 open water season.

In spite of the well-documented capacity of various chemical dispersants to reduce the size of oil particles and disperse them in the water column, we are unaware of any study where dispersants were shown to enhance in situ biodegradation. Corexit 9527 severely reduced the mineralization of ^{14}C-hexadecane across 60 days of incubation in laboratory experiments. Similar observations have been made from water samples collected at Frobisher Bay, N.W.T. (unpublished data). There was an extended lag before hexadecane mineralization occurred where Corexit was present. This lag was reduced in samples supplemented with nitrate and phosphate. Similar observations have been made by Foght and Westlake (1982). We agree with their suggestion that an additional carbon input by means of Corexit may stress nitrate-phosphate levels and delay oxidation of hydrocarbons. In fact, components of the dispersant could be preferentially utilized by bacteria over hydrocarbon components of petroleum including hexadecane. By this hypothesis, biodegradation of petroleum in a marine environment low in levels of nitrate and phosphate would be severely retarded by the addition of a dispersant. The preferential oxidation of the dispersant by bacterial floras would deplete nutrient levels thereby retarding oxidation of hydrocarbons.

In 1982, observations of bacterial numbers and activity in the water column and sediments of the bays have been continued. Although collections were made in the water column, emphasis was placed on the oiled sediments of bays 9, 10 and 11. The results of these studies will be the subject of another report.

6.0 CONCLUSIONS

1. At the concentrations of oil released into the water by the surface and dispersed oil releases, the effect on measured bacterial activity was transient and minimal.
2. No changes in bacterial activity in the sediments could be ascribed to either oil release.
3. Counts of bacterial cells by epifluorescent microscopy or colonies by a plating technique were not affected by the oil releases.
4. Corexit 9527 significantly altered kinetic parameters of glutamic acid uptake in laboratory experiments. This effect could not be determined during the dispersed release where the concentration of Corexit was three orders of magnitude lower than in the laboratory experiments.
5. An oil-degrading capacity was determined in the water and sediments of all bays. This capacity did not appear to be altered by the oil releases.

7.0 REFERENCES

Alexander, S. K. and J. R. Schwarz. 1980. Short-term effects of South Louisiana and Kuwait crude oils on glucose utilization by marine bacterial populations. Appl. Environ. Microbiol. 38: 78-83.

Boehm, P. D., D. L. Fiest and P. Hirtzer. 1982. Chemistry: 2. Analytical biogeochemistry--1981 study results. Baffin Island Oil Spill (BIOS) project working report 81-2: 354 p. Environmental Protection Service, Canadian Department of the Environment, Edmonton, Alberta.

Bunch, J. N., C. Bédard and T. Cartier. 1983. Chemistry: 1. Environmental chemistry--1981 study results. Baffin Island Oil Spill (BIOS) project working report 81-1: in preparation.

Bunch, J. N., R. C. Harland and J. Lalibertê. 1981. Abundance and activity of heterotrophic marine bacteria in selected bays at Cape Hatt, N.W.T. 1980. Can. MS Rep. Fish. Aquat. Sci. 1611: xiii + 68 p.

Cross, W. E. and D. H. Thomson. 1982. Macrobenthos--1981 study results. Baffin Island Oil Spill (BIOS) project working report 81-3: 105 p. Environmental Protection Service, Canadian Department of the Environment, Edmonton, Alberta.

Dickins, D. F. 1982a. Discharge systems--1981 study results. Baffin Island Oil Spill (BIOS) project working report 81-9: 62 p. Environmental Protection Service, Canadian Department of the Environment, Edmonton, Alberta.

Dickins, D. F. 1982b. Ice conditions--1981 study results. Baffin Island Oil Spill (BIOS) project working report 81-8: 41 p. Environmental Protection Service, Canadian Department of the Environment, Edmonton, Alberta.

Dowd, J. E. and D. S. Riggs. 1965. A comparison of estimates of Michaelis-Menten kinetic constants from various linear transformations. J. Biol. Chem. 240: 863-869.

Foght, J. M. and D. W. S. Westlake. 1982. Effect of the dispersant Corexit 9527 on the microbial degradation of Prudhoe Bay oil. Can. J. Microbiol. 28: 117-122.

Griffiths, R. P., T. M. McNamara, B. A. Caldwell and R. Y. Morita. 1981. Field observations on the acute effects of crude oil on glucose and glutamate uptake in samples collected from arctic and subarctic waters. Appl. Environ. Microbiol. 41: 1400-1406.

Helwig, J. T. and K. A. Council. 1979. SAS user's guide. 494 p. SAS Institute Inc., Raleigh, North Carolina.

Hodson, R. E., F. Azam and R. F. Lee. 1977. Effects of four oils on marine bacterial populations: controlled ecosystem pollution experiment. Bull. Mar. Sci. 27: 119-126.

Parsons, T. R., M. Takahashi and B. Hargrave. 1977. Biological oceanographic processes. Second edition. 332 p. Pergamon Press, Oxford.

Sokal, R. R. and F. J. Rohlf. 1969. Biometry. The principles and practice of statistics in biological research. 776 p. W. H. Freeman and Company, San Francisco.

8.0 Figures

Figures 1-14

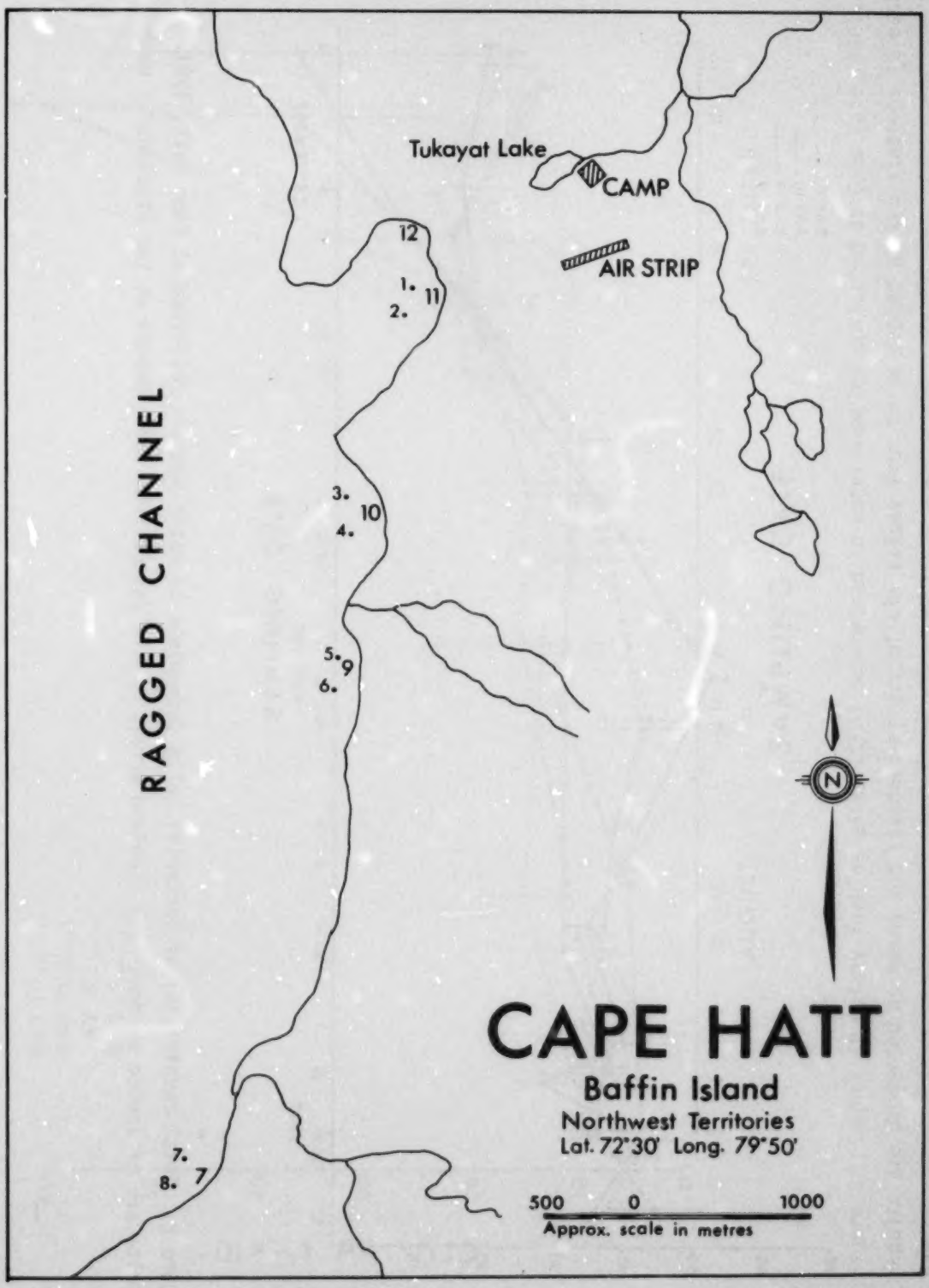


Figure 1. Microbiological stations occupied at Cape Hatt during 1981.

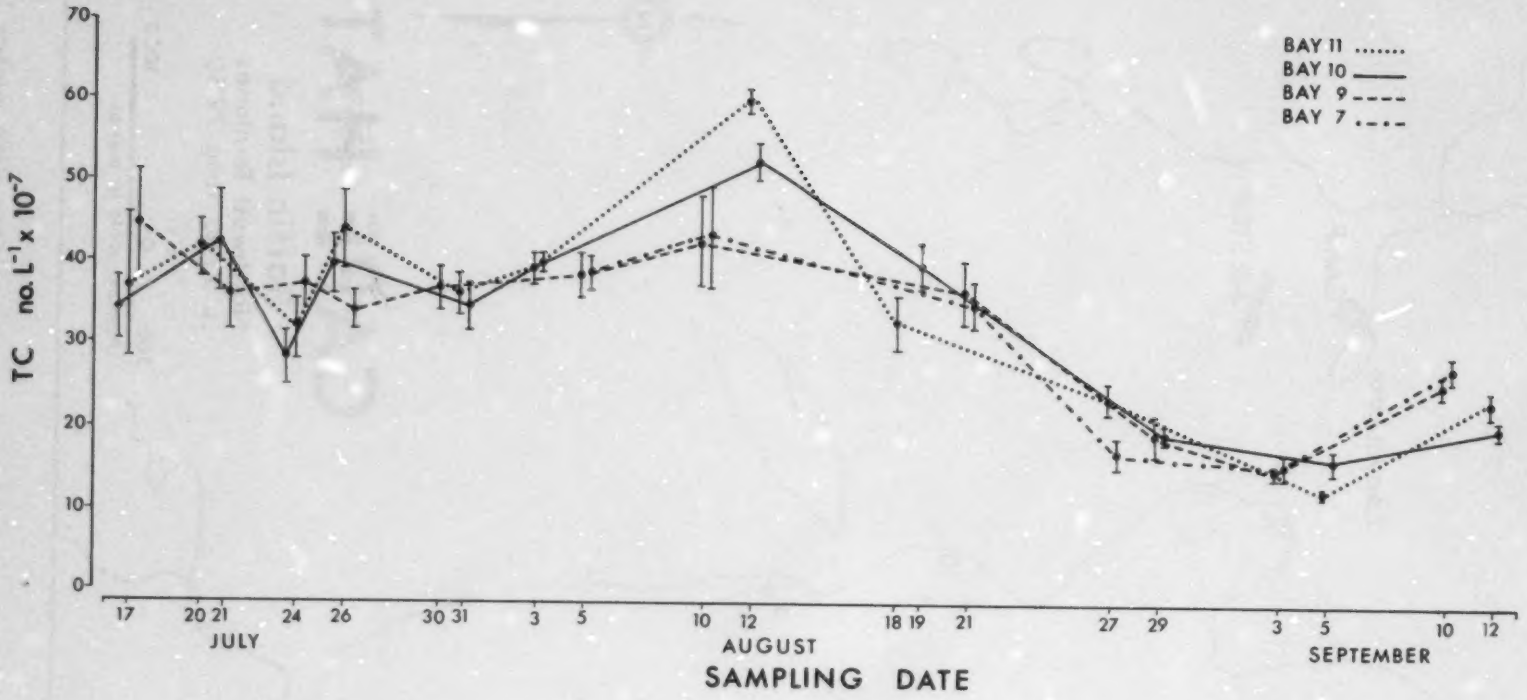


Figure 2. Total counts (TC) of bacterial cells determined in water samples collected at Cape Hatt, 1981. Results are presented as means and standard errors of six values from three depths at two stations in each bay.

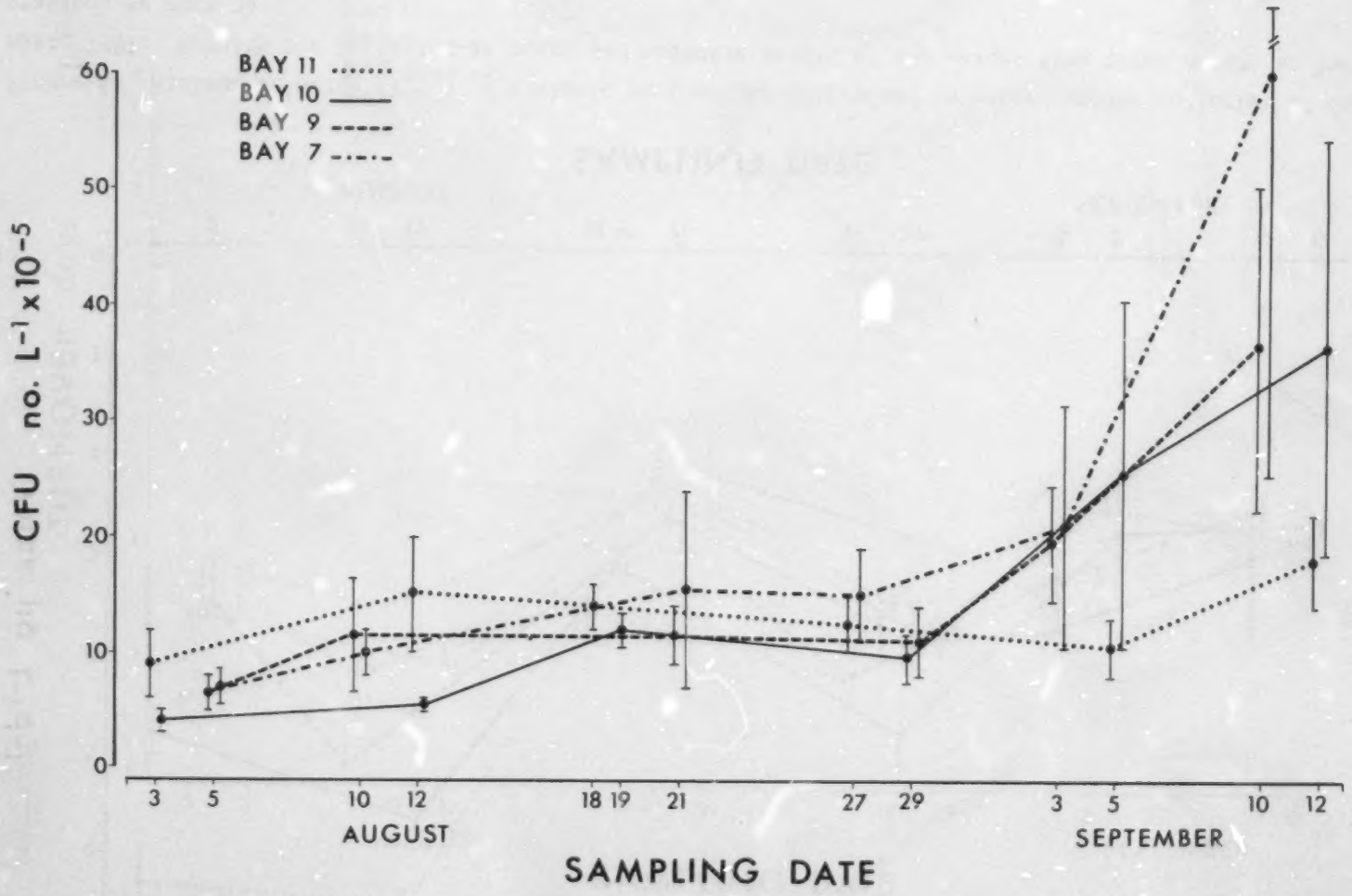


Figure 3. Counts of colony-forming units (CFU) determined in water samples collected at Cape Hatt, 1981. Results are presented as means and standard errors of six values from three depths at two stations in each bay.

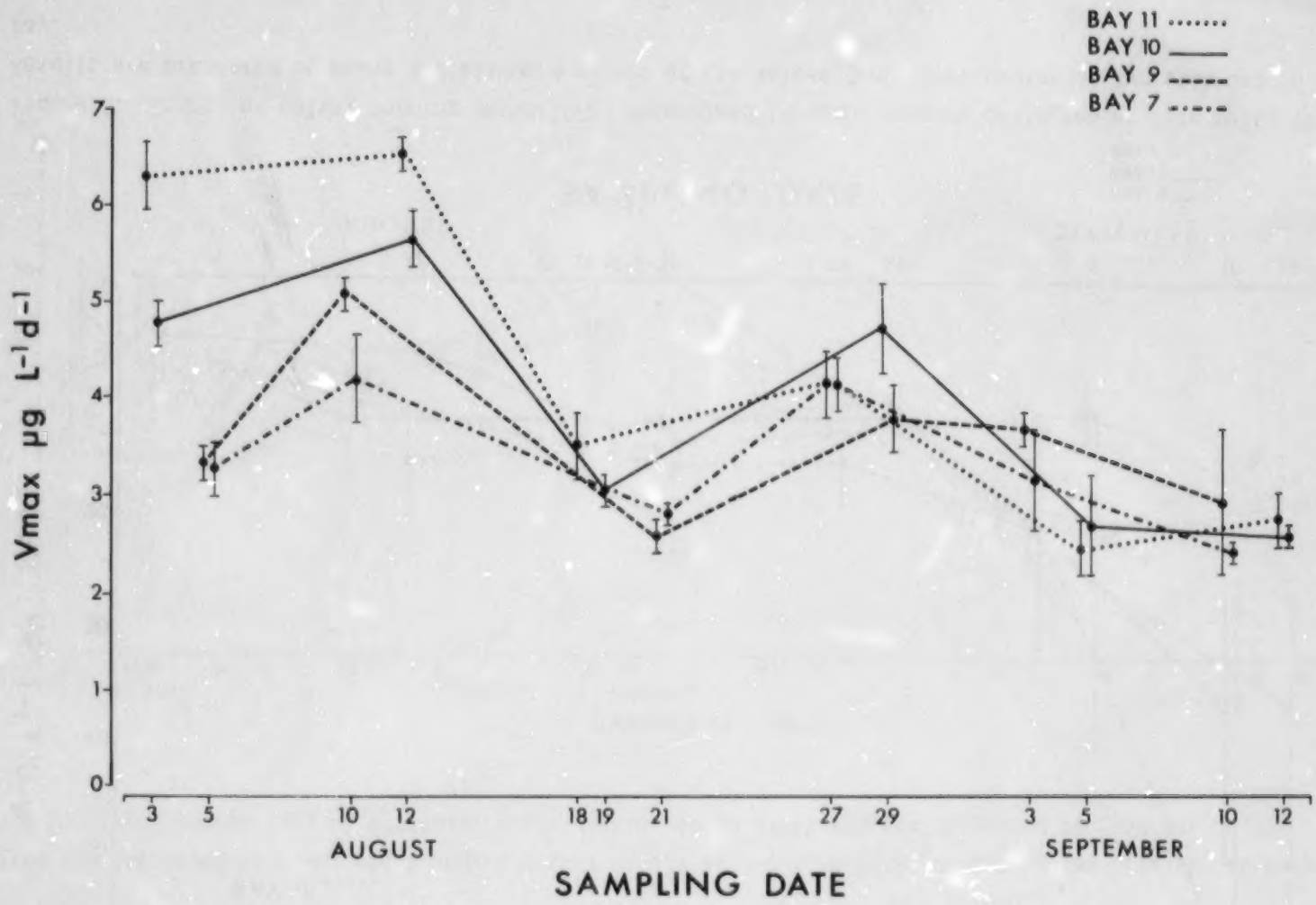


Figure 4. Maximum velocity (V_{max}) of glutamic acid uptake determined in water samples collected at Cape Hatt, 1981. Results are presented as means and standard errors of six values from three depths at two stations in each bay.

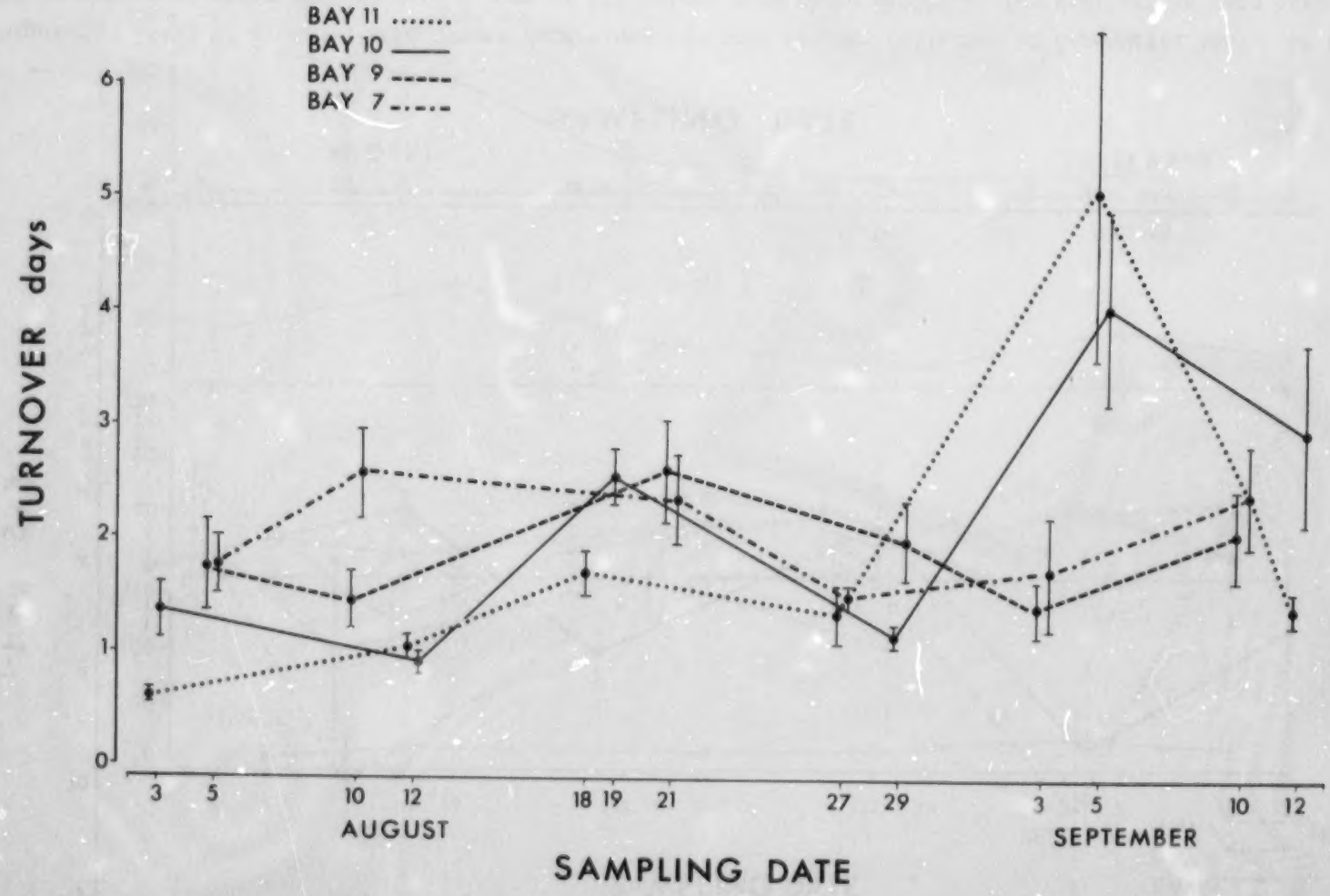


Figure 5. Turnover of glutamic acid uptake determined in water samples collected at Cape Hatt, 1981. Results are presented as means and standard errors of six values from three depths at two stations in each bay.

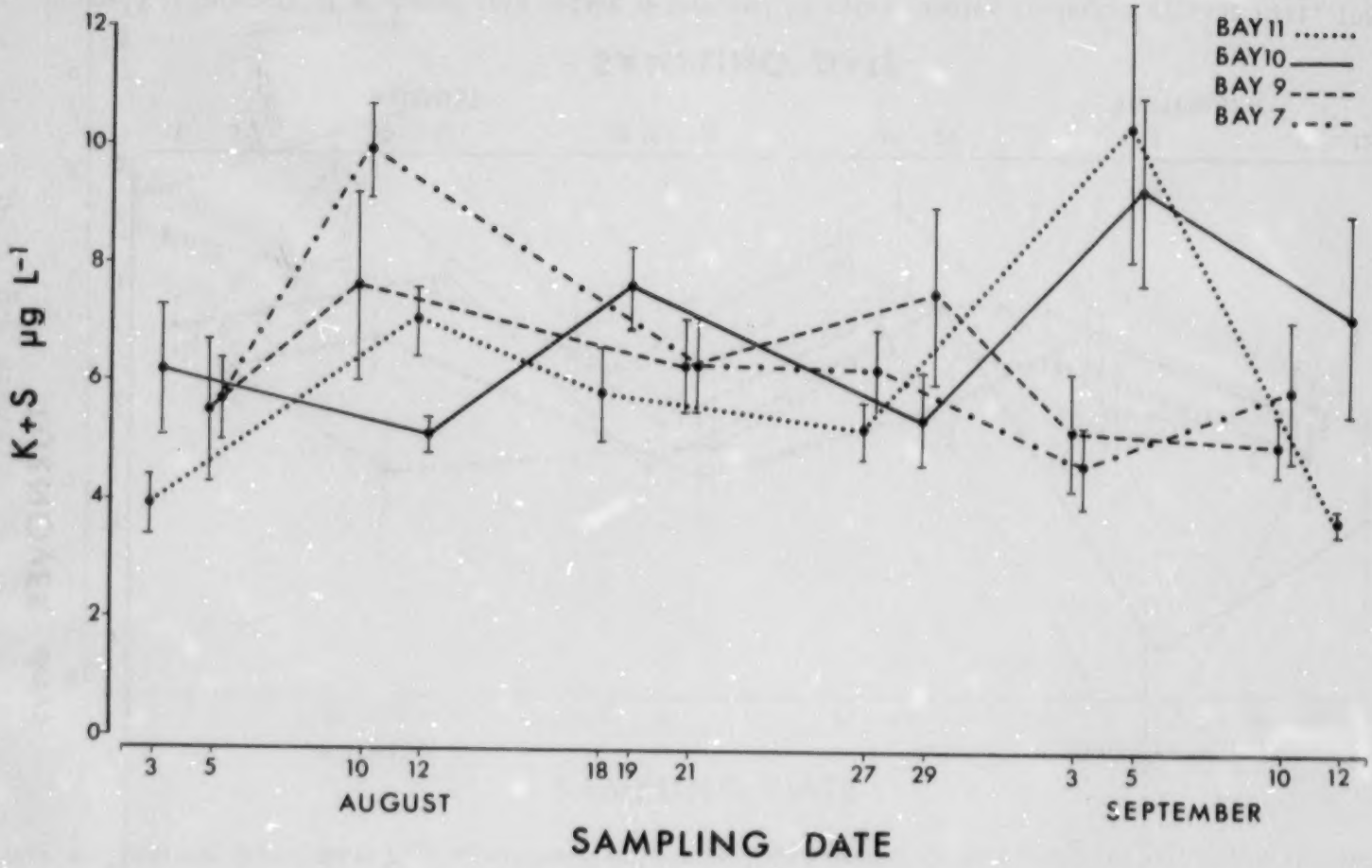


Figure 6. (K+S) of glutamic acid uptake determined in water samples collected at Cape Hatt, 1981. Results are presented as means and standard errors of six values from three depths at two stations in each bay.

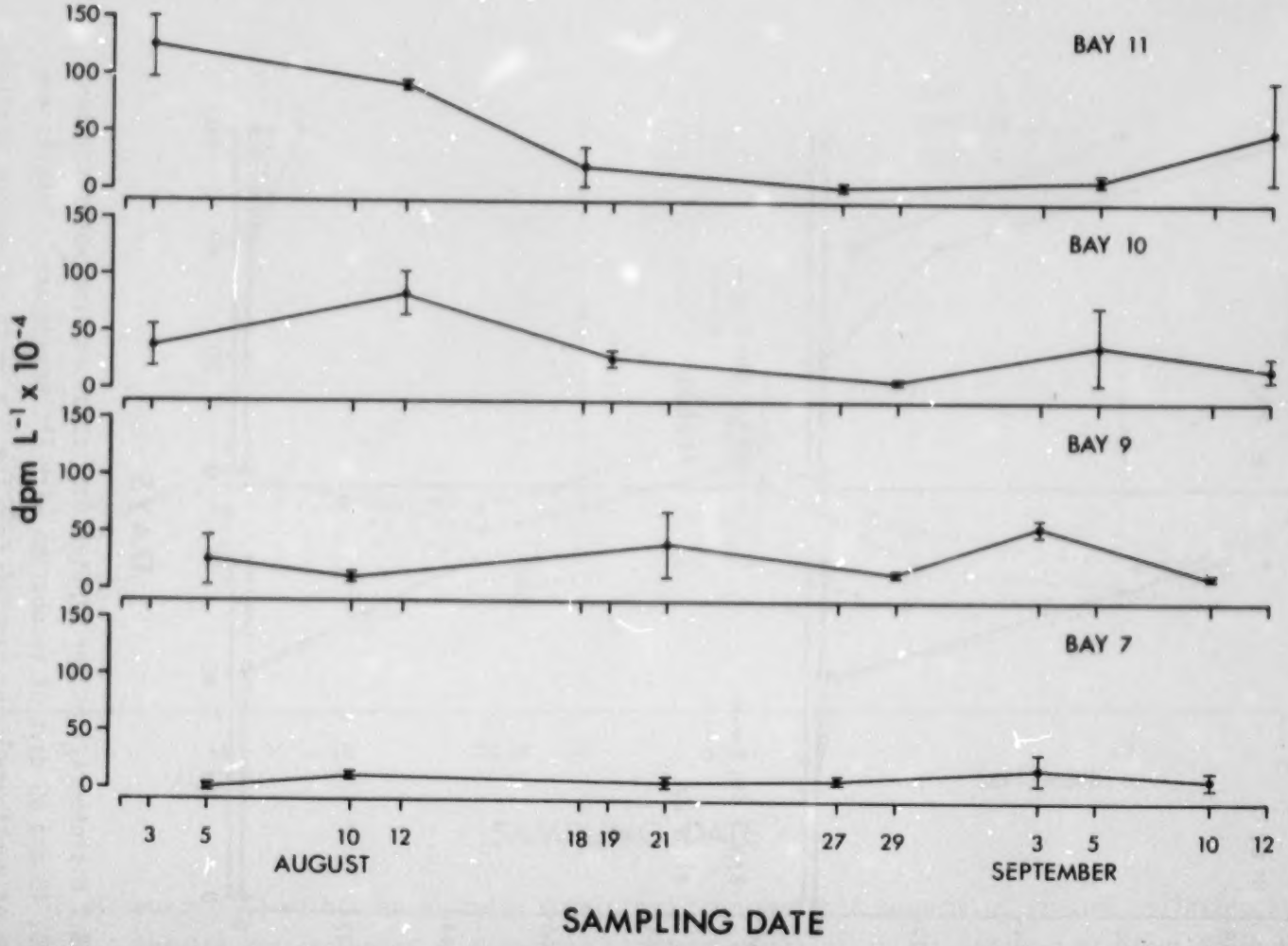


Figure 7. Disintegrations per minute (dpm) obtained from water samples after 60 days incubation with ^{14}C-hexadecane. Results are the means and standard errors of replicate water samples from the 5 m depth of two stations in each bay. All incubations were supplemented with inorganic nutrients.

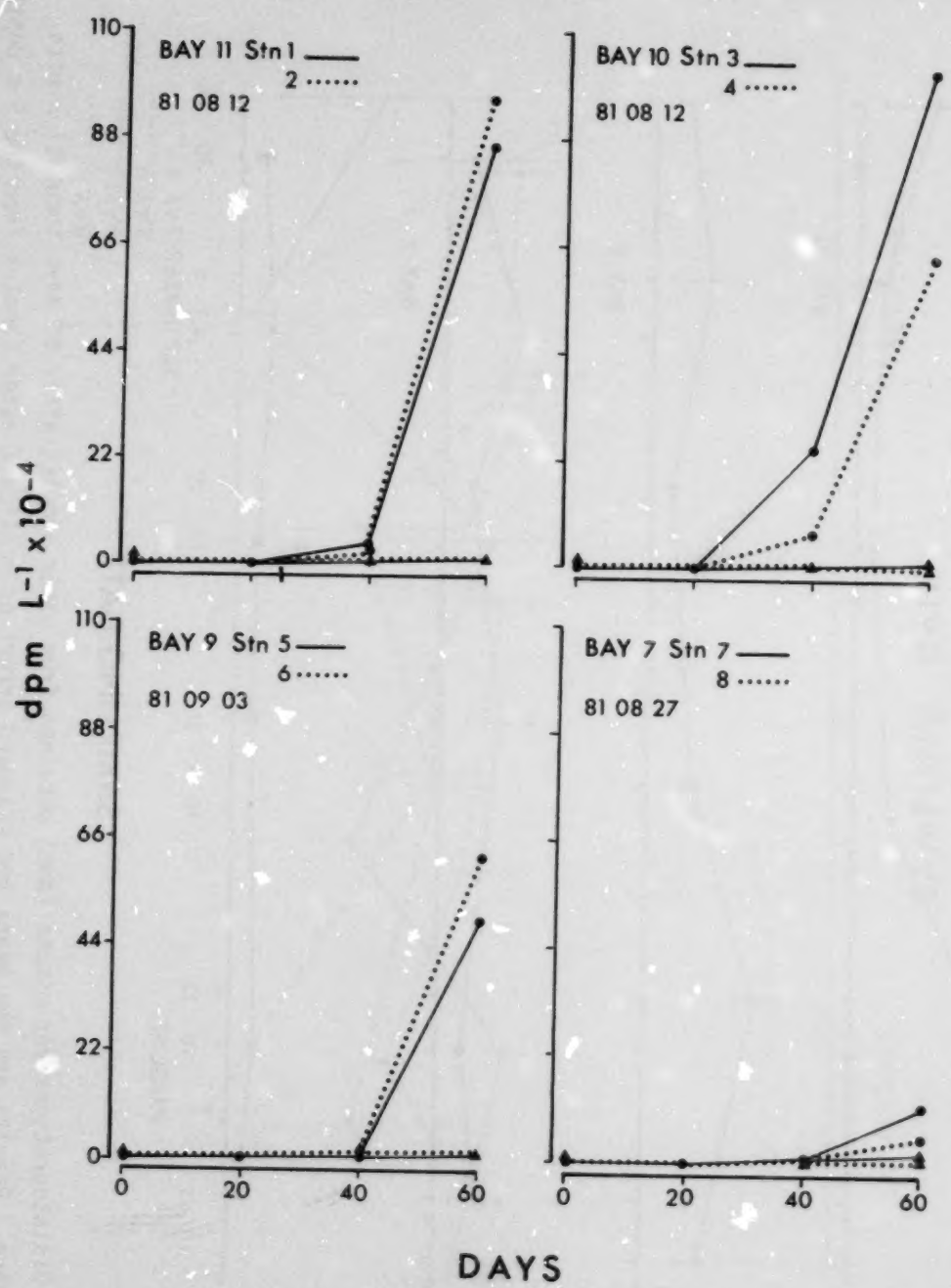


Figure 8. Disintegrations per minute (dpm) obtained from water samples after 20, 40 and 60 days of incubation with ^{14}C-hexadecane. Results are the mean of duplicate water samples from the 5 m depth of various stations. All incubations were supplemented with inorganic nutrients. Similar samples supplemented with Corexit 9527 were also incubated for 40 and 60 days (•) hexadecane, (▲) hexadecane + Corexit.

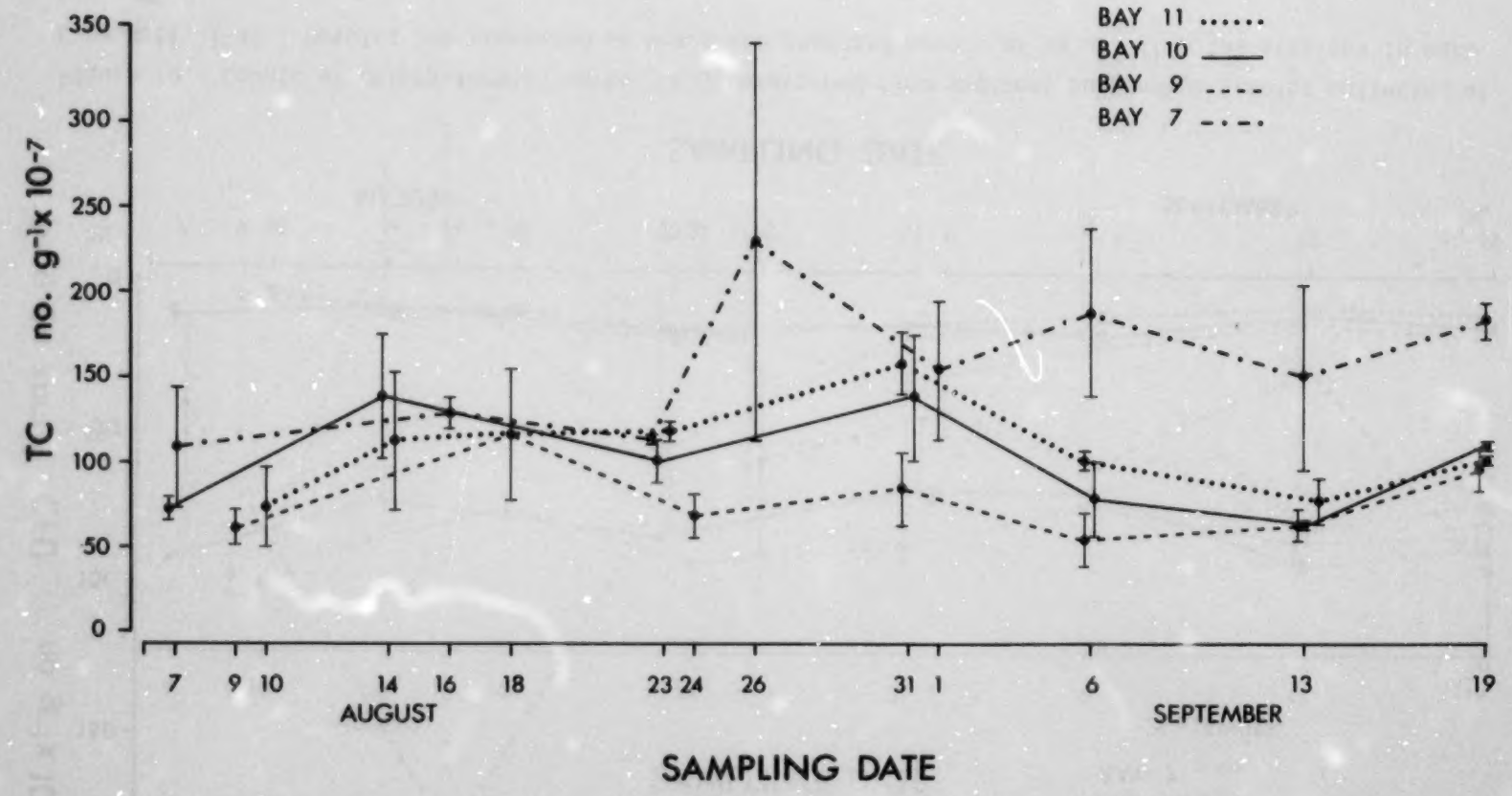


Figure 9. Total counts (TC) of bacterial cells determined from sediment suspension samples collected at Cape Hatt, 1981. Results are presented as means and standard errors of values from two stations in each bay.

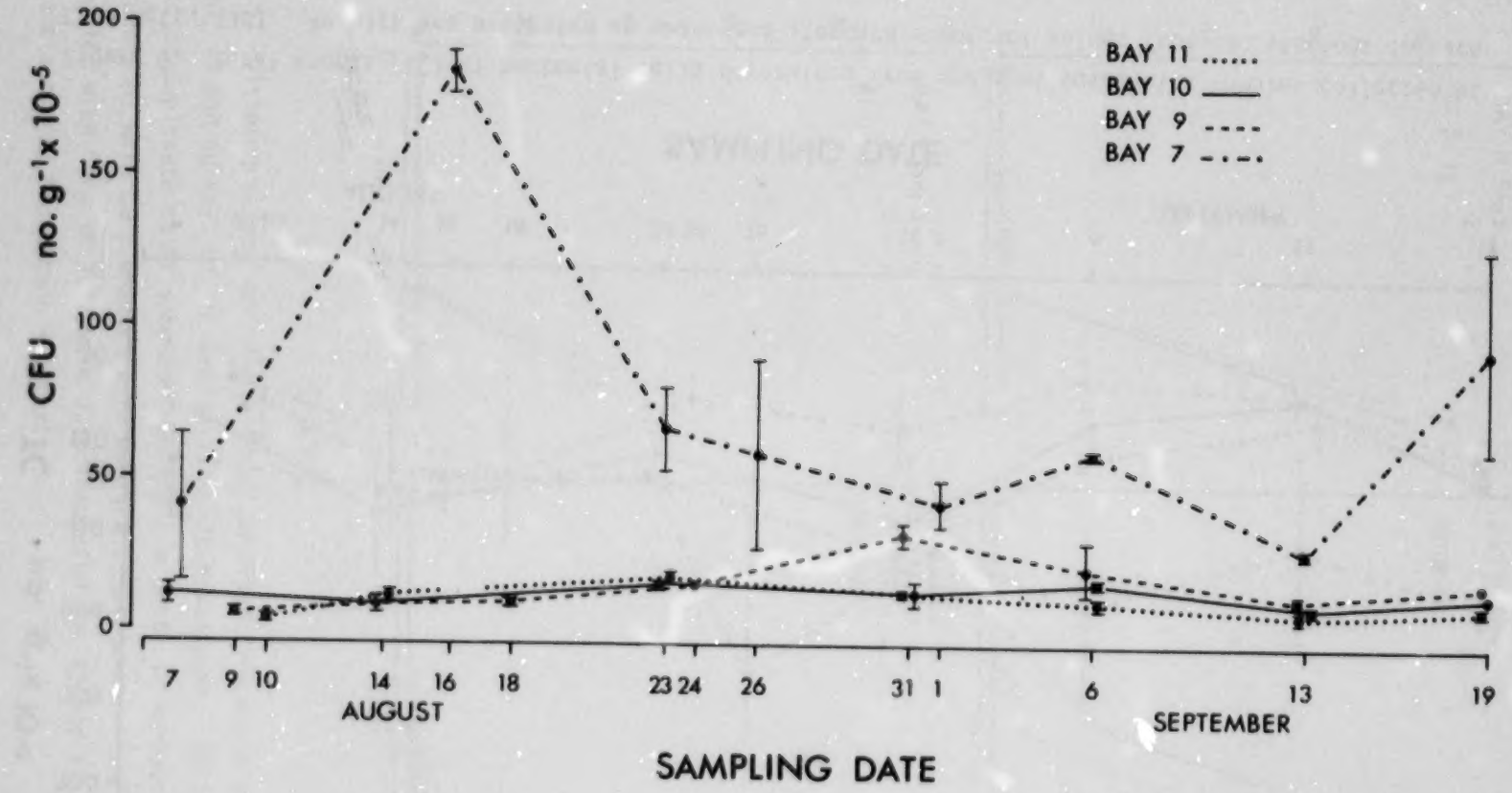


Figure 10. Counts of colony-forming units (CFU) determined from sediment suspension samples collected at Cape Hatt, 1981. Results are presented as means and standard errors of values from two stations in each bay.

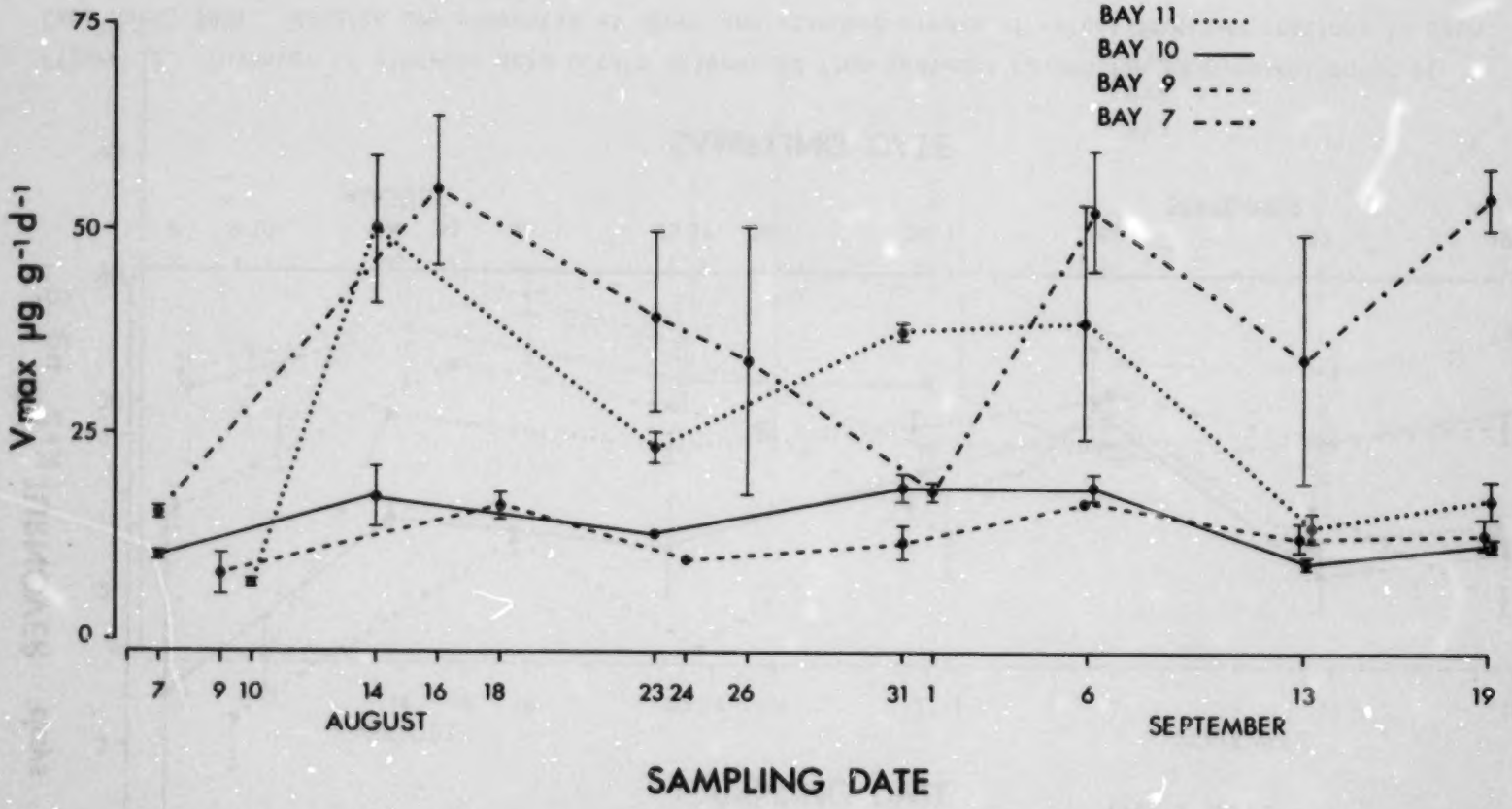


Figure 11. Maximum velocity (V_{max}) of glutamic acid uptake determined from sediment suspension samples collected at Cape Hatt, 1981. Results are presented as means and standard errors of values from two stations in each bay.

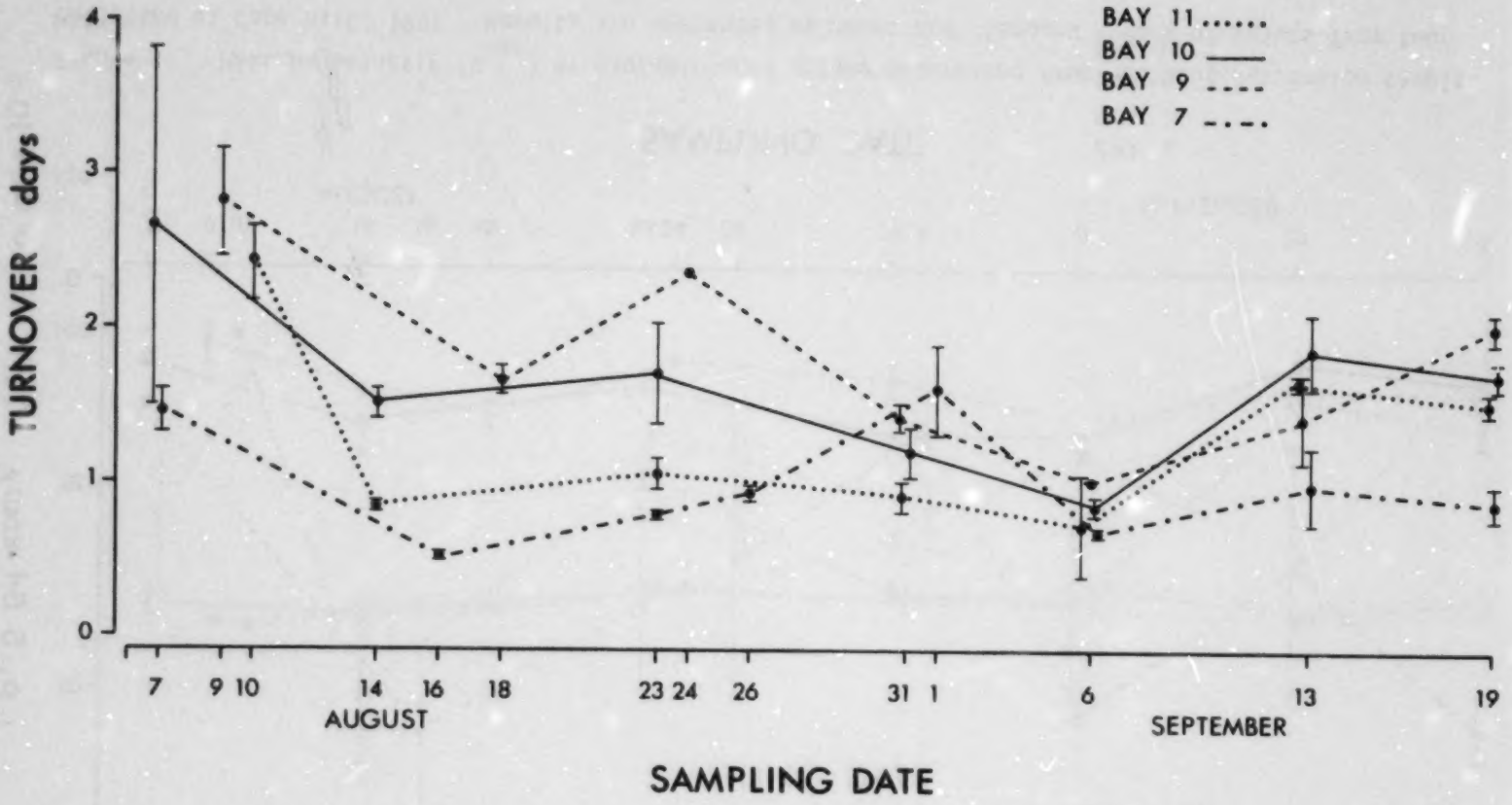


Figure 12. Turnover of glutamic acid uptake determined from sediment suspension samples collected at Cape Hatt, 1981. Results are presented as means and standard errors of values from two stations in each bay.

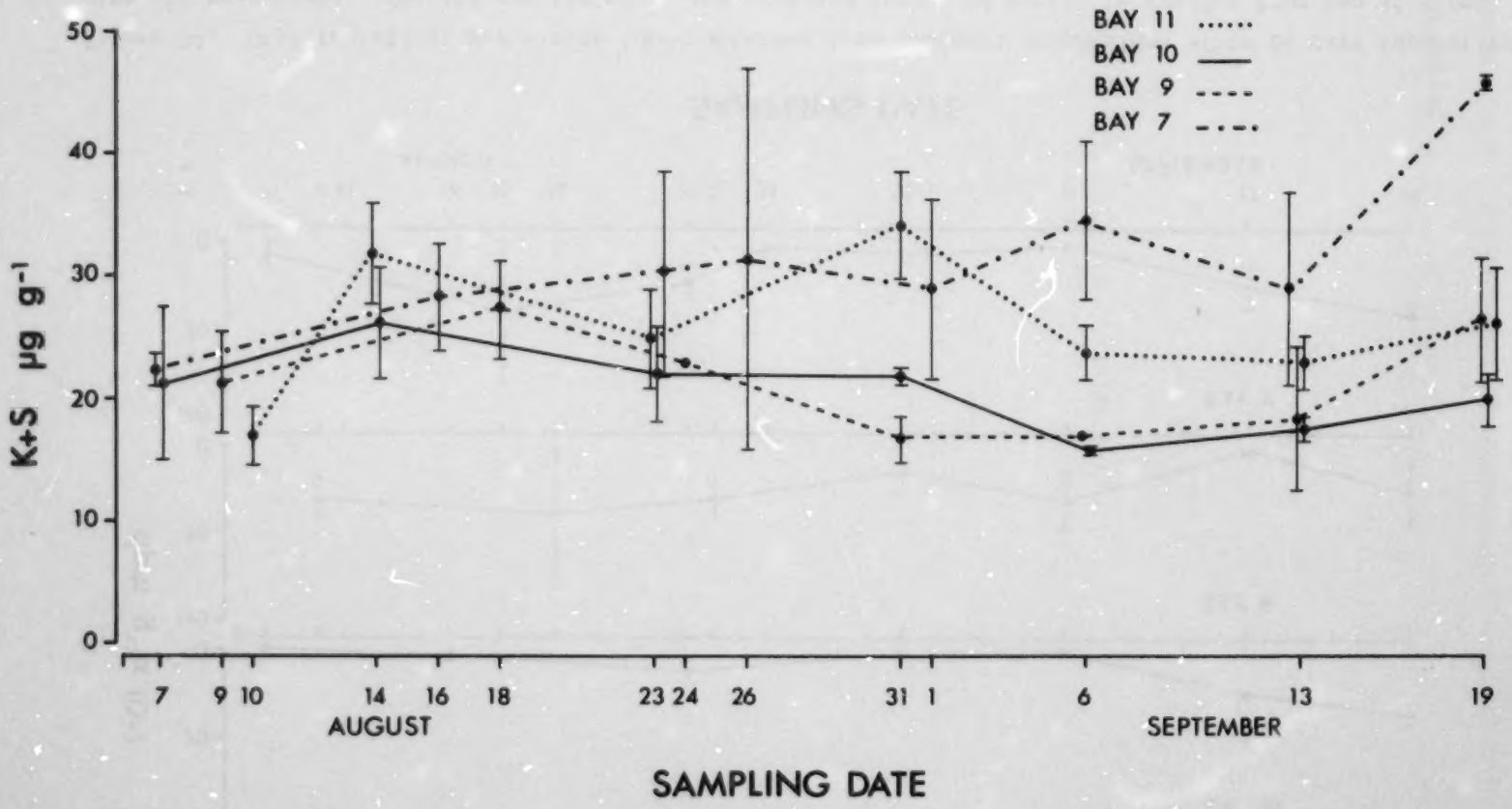


Figure 13. (K+S) of glutamic acid uptake determined from sediment suspension samples collected at Cape Hatt, 1981. Results are presented as means and standard errors of values from two stations in each bay.

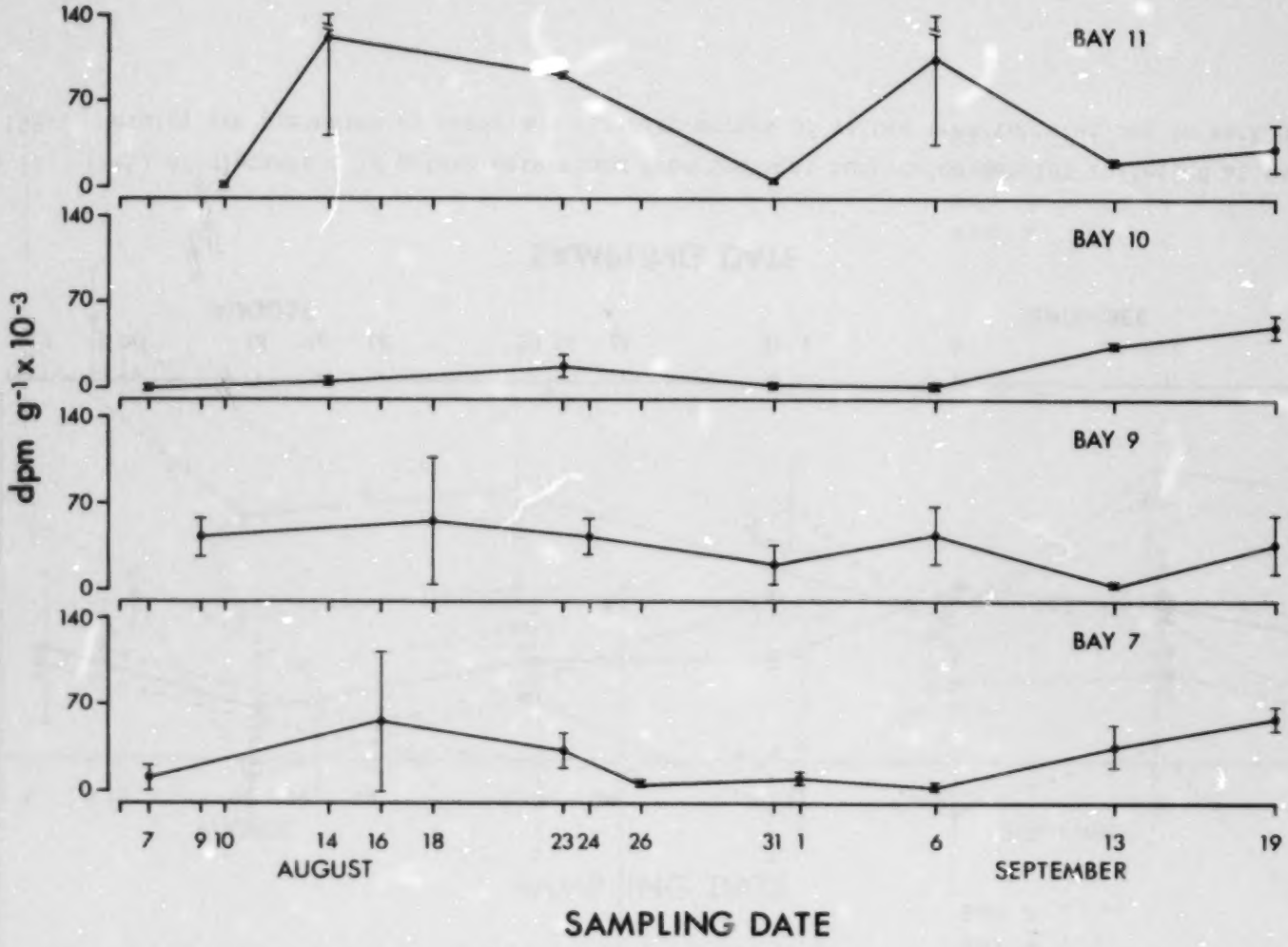


Figure 14. Disintegrations per minute (dpm) obtained from sediment suspensions after 60 days incubation with ^{14}C-hexadecane. Results are the means and standard errors of replicate samples from two stations in each bay and are expressed as dpm per gram dry weight of sediment. All incubations were supplemented with inorganic nutrients.

9.0 Tables

Tables 1-13

Table 1. Water column station occupations at Cape Hatt, 1981.

Cycle	Date	Bay	Stations
1	08 03	11	1 2
		10	3 4
	08 05	9	5 6
		7	7 8
2	08 10	9	5 6
		7	7 8
	08 12	11	1 2
		10	3 4
3	08 18	11	1 2
	08 19	10	3 4
	08 21	9	5 6
		7	7 8
4	08 27	11	1 2
		7	7 8
	08 29	10	3 4
		9	5 6
5	09 03	9	5 6
		7	7 8
	09 05	11	1 2
		10	3 4
6	09 10	9	5 6
		7	7 8
	09 12	11	1 2
		10	3 4

Table 2. Sediment station occupations at Cape Hatt, 1981.

Cycle	Date	Bay	Stations
1	08 07	10	3 4
		7	7 8
	08 09	9	5 6
	08 10	11	1 2
2	08 14	11	1 2
		10	3 4
	08 16	7	7 8
	08 18	9	5 6
3	08 23	11	1 2
		10	3 4
		7	7 8
	08 24	9	5 6
	08 26	7	7 8
4	08 31	11	1 2
		10	3 4
		9	5 6
	09 01	7	7 8
5	09 06	11	1 2
		10	3 4
		9	5 6
		7	7 8
6	09 13	11	1 2
		10	3 4
		9	5 6
		7	7 8

Table 2 (cont'd)

Cycle	Date	Bay	Stations
7	09 19	11	1 2
		10	3 4
		9	5 6
		7	7 8

Table 3. Determinations of total counts (TC) of bacterial cells from water samples collected at Cape Hatt during July 1981.

Station	Date	Depth	TC
		m	no. L^{-1} (10^{-7})
1	07 17	0	77.36
		5	26.10
		10	47.31
2	07 17	0	19.38
		5	31.84
		10	20.78
3	07 17	0	28.54
		5	32.16
		10	28.18
4	07 17	0	27.58
		5	51.50
		9	39.76
5	07 17	0	30.63
		5	34.77
		10	36.50
6	07 17	0	44.30
		5	46.67
		10	75.86
1	07 20	0	32.52
		5	53.53
		10	46.69
2	07 20	0	34.27
		5	41.67
		10	44.48
3	07 21	0	29.77
		5	51.60
		9	62.29
4	07 21	0	33.47
		5	25.79
		10	53.91

Table 3 (cont'd)

Station	Date	Depth	TC
		m	no. L^{-1} (10^{-7})
5	07 21	0	27.46
		5	37.95
		10	53.99
6	07 21	0	23.40
		5	42.41
		9	33.04
1	07 24	0	27.26
		5	39.60
		10	32.68
2	07 24	0	17.39
		5	41.75
		10	33.39
3	07 24	0	17.57
		5	39.28
		10	28.09
4	07 24	0	20.59
		5	34.89
		10	30.47
5	07 24	0	30.20
		5	29.40
		10	36.43
6	07 24	0	33.92
		5	44.49
		10	49.88
1	07 26	0	42.55
		5	54.15
		10	41.39
2	07 26	0	58.71
		5	40.62
		10	27.92

Table 3 (cont'd)

Station	Date	Depth m	TC no. L^{-1} (10^{-7})
3	07 26	0	42.63
		5	29.20
		10	38.47
4	07 26	0	55.88
		5	38.63
		10	34.67
5	07 26	0	31.62
		5	31.46
		10	31.86
6	07 26	0	36.55
		5	45.19
		10	29.71
1	07 31	0	27.44
		5	29.95
		10	37.75
2	07 31	0	38.29
		5	42.48
		10	41.74
3	07 31	0	34.27
		5	38.56
		10	24.15
4	07 31	0	40.70
		5	42.28
		10	28.61
5	07 30	0	38.46
		5	34.97
		10	32.66
6	07 30	0	44.59
		5	43.42
		10	27.34

Table 4. Determinations of colony-forming units (CFU), total counts (TC) of bacterial cells, maximum velocity (V_{max}), turnover (T) and (K+S) (see text) of glutamic acid uptake from water samples collected at Cape Hatt during August and September 1981.

Station	Date	Depth	CFU	TC	V_{max}	T	(K+S)
		m	no. L^{-1} (10^{-5})	no. L^{-1} (10^{-7})	µg L^{-1} d^{-1}	d	µg L^{-1}
1	08 03	0	23.80	44.86	4.96	0.75	3.73
		5	4.20	44.76	6.47	0.47	3.04
		10	9.20	39.43	6.61	0.90	5.93
2	08 03	0	6.50	32.93	5.94	0.47	2.79
		5	5.90	36.75	6.48	0.60	3.87
		10	4.40	37.19	7.46	0.55	4.07
3	08 03	0	7.70	36.62	3.93	1.29	5.06
		5	3.10	41.64	5.32	1.14	6.06
		10	5.00	36.85	5.61	0.82	4.61
4	08 03	0	3.90	40.37	4.59	0.65	2.98
		5	2.40	40.10	4.77	1.76	8.38
		10	3.10	45.06	4.36	2.33	10.15
5	08 05	0	12.60	31.34	3.37	0.57	1.93
		5	4.20	45.71	3.04	2.76	8.40
		10	4.80	43.37	4.08	0.81	3.31
6	08 05	0	8.20	40.92	2.86	1.37	3.92
		5	3.10	29.90	3.43	2.00	6.85
		10	6.70	39.81	3.10	2.84	8.80

Table 4 (cont'd)

Station	Date	Depth	CFU	TC	Vmax	T	(K+S)
		m	no. L^{-1} (10^{-5})	no. L^{-1} (10^{-7})	µg L^{-1} d^{-1}	d	µg L^{-1}
7	08 05	0	6.10	43.37	3.48	0.78	2.71
		5	4.70	44.61	3.75	1.73	6.48
		10	9.40	40.02	3.05	2.37	7.21
8	08 05	0	12.90	32.11	2.27	1.94	4.40
		5	1.80	33.11	4.34	1.61	7.00
		10	6.10	39.58	2.79	2.20	6.12
1	08 12	0	32.70	54.66	6.66	1.36	9.04
		5	6.70	64.94	6.20	1.12	6.96
		10	24.90	58.64	5.96	1.22	7.26
2	08 12	0	4.50	59.01	6.90	1.01	6.94
		5	4.60	63.37	6.88	1.05	7.20
		10	16.90	59.81	6.50	0.68	4.43
3	08 12	0	5.40	48.93	5.96	0.68	4.08
		5	6.50	52.11	5.91	0.94	5.53
		10	3.80	61.02	6.53	0.79	5.14
4	08 12	0	5.90	45.81	5.10	1.15	5.85
		5	4.90	51.67	4.80	1.10	5.29
		10	6.60	56.40	5.69	0.80	4.55
5	08 10	0	4.90	42.87	4.56	1.15	5.22
		5	1.70	29.61	4.73	1.09	5.17
		10	7.90	48.46	5.62	1.66	9.33

Table 4 (cont'd)

Station	Date	Depth	CFU	TC	V_{max}	T	(K+S)
		m	no. L^{-1} (10^{-5})	no. L^{-1} (10^{-7})	$\mu g\ L^{-1}\ d^{-1}$	d	$\mu g\ l^{-1}$
6	08 10	0	9.70	33.06	4.93	1.23	6.07
		5	11.50	36.72	4.95	1.03	5.09
		10	34.50	66.91	5.76	2.56	14.75
7	08 10	0	6.00	30.33	4.65	1.54	7.17
		5	10.50	39.94	4.74	1.73	8.19
		10	10.40	47.42	5.66	1.75	9.89
8	08 10	0	12.90	27.28	4.27	2.79	11.91
		5	3.30	44.24	2.83	3.81	10.81
		10	18.20	70.61	3.07	3.66	11.25
1	08 18	0	23.70	43.72	4.05	2.28	9.21
		5	9.30	40.74	3.83	1.72	6.58
		10	14.70	35.84	2.74	1.77	4.86
2	08 18	0	14.10	25.31	4.15	1.29	5.34
		5	14.00	25.99	4.13	0.89	3.67
		10	8.70	26.55	2.15	2.28	4.90
3	08 19	0	15.30	34.95	3.06	1.91	5.84
		5	11.90	36.97	2.84	2.70	7.68
		10	9.30	32.14	2.64	2.68	7.09
4	08 19	0	7.00	49.55	3.70	1.70	6.28
		5	18.10	37.73	3.33	3.14	10.43
		10	10.80	46.57	2.66	3.14	8.35

Table 4 (cont'd)

Station	Date	Depth	CFU	TC	V_{max}	T	(K+S)
		m	no. L^{-1} (10^{-5})	no. L^{-1} (10^{-7})	$\mu g\ L^{-1}\ d^{-1}$	d	$\mu g\ L^{-1}$
5	08 21	0	20.60	28.88	2.68	3.38	9.06
		5	6.80	44.78	2.68	1.91	5.12
		10	4.20	28.78	1.80	4.27	7.66
6	08 21	0	12.20	30.23	2.43	2.76	6.71
		5	7.10	52.38	2.66	2.00	5.33
		10	17.40	34.89	2.96	1.30	3.84
7	08 21	0	55.50	41.16	3.08	2.13	6.56
		5	11.00	44.14	3.24	1.15	3.71
		10	5.00	28.14	2.76	2.67	7.37
8	08 21	0	14.00	38.31	2.74	1.61	4.42
		5	2.10	33.08	2.26	4.07	9.19
		10	3.40	26.69	2.72	2.45	6.65
1	08 27	0	8.40	30.99	4.20	1.26	5.30
		5	15.40	25.89	3.07	1.77	5.44
		10	22.90	19.50	3.30	2.30	7.58
2	08 27	0	15.80	21.15	5.37	0.89	4.78
		5	6.60	19.87	4.22	1.10	4.63
		10	6.40	26.25	4.59	0.79	3.64
3	08 29	0	15.30	19.46	3.54	1.10	3.88
		5	9.00	17.85	4.41	0.95	4.17
		10	13.20	18.25	5.97	1.21	7.22

Table 4 (cont'd)

Station	Date	Depth	CFU	TC	V_{max}	T	(K+S)
		m	no. L^{-1} (10^{-5})	no. L^{-1} (10^{-7})	$\mu g\ L^{-1}\ d^{-1}$	d	$\mu g\ L^{-1}$
4	08 29	0	10.30	24.36	3.68	0.87	3.20
		5	5.00	21.59	4.30	1.33	5.71
		10	3.50	16.50	6.35	1.29	8.22
5	08 29	0	17.10	16.73	4.68	1.75	8.20
		5	18.20	29.65	4.37	1.95	8.54
		10	3.30	12.59	4.04	3.48	14.05
6	08 29	0	6.80	23.40	3.42	1.06	3.64
		5	4.70	15.63	3.89	1.23	4.79
		10	15.40	17.67	2.45	2.43	5.96
7	08 27	0	8.50	22.15	3.82	1.47	5.63
		5	8.90	12.79	2.87	1.53	4.39
		10	29.50	14.20	3.78	1.58	5.96
8	08 27	0	17.50	23.04	4.72	2.03	9.58
		5	20.70	17.44	4.53	1.24	5.62
		10	4.10	12.91	4.94	1.17	5.79
1	09 05	0	11.10	14.83	2.18	5.19	11.31
		5	6.90	13.94	1.82	6.05	11.00
		10	19.10	12.68	1.57	10.30	16.17
2	09 05	0	0.70	12.73	3.62	1.20	4.35
		5	14.70	10.21	3.02	1.08	3.25
		10	11.30	11.24	2.36	6.56	15.48

Table 4 (cont'd)

Station	Date	Depth	CFU	TC	V_{max}	T	(K+S)
		m	no. L^{-1} (10^{-5})	no. L^{-1} (10^{-7})	$\mu g\ L^{-1}\ d^{-1}$	d	$\mu g\ L^{-1}$
3	09 05	0	1.20	14.69	2.10	7.73	16.21
		5	20.80	23.65	3.47	3.49	12.11
		10	99.10	14.66	5.13	1.18	6.04
4	09 05	0	14.90	14.15	1.93	3.39	6.55
		5	13.00	15.63	1.87	4.13	7.72
		10	3.90	15.80	1.68	4.28	7.17
5	09 03	0	20.80	14.17	3.49	1.93	6.74
		5	13.20	13.54	3.83	2.28	8.75
		10	41.60	12.39	3.39	0.78	2.63
6	09 03	0	18.70	15.65	4.44	1.16	5.14
		5	6.40	17.31	3.84	1.39	5.35
		10	16.20	16.86	3.22	0.84	2.71
7	09 03	0	1.00	17.91	2.59	1.25	3.24
		5	11.60	9.40	3.91	1.31	5.14
		10	23.60	16.86	3.20	1.38	4.43
8	09 03	0	69.60	13.87	5.10	1.00	5.11
		5	15.00	17.59	2.17	1.19	2.58
		10	3.90	18.19	1.74	4.20	7.33
1	09 12	0	32.20	22.74	3.40	1.04	3.54
		5	12.60	19.45	2.09	1.60	3.34
		10	9.00	30.53	2.14	2.07	4.41

Table 4 (cont'd)

Station	Date	Depth	CFU	TC	V_{max}	T	(K+S)
		m	no. L^{-1} (10^{-5})	no. L^{-1} (10^{-7})	$\mu g\ L^{-1}\ d^{-1}$	d	$\mu g\ L^{-1}$
2	09 12	0	25.00	23.97	3.18	1.21	3.85
		5	6.10	24.70	3.67	0.99	3.64
		10	22.20	20.53	1.97	1.59	3.13
3	09 12	0	28.70	24.20	2.69	1.61	4.32
		5	10.30	18.62	2.74	1.16	3.18
		10	32.10	19.85	2.49	1.21	3.01
4	09 12	0	2.90	19.45	2.95	3.37	9.94
		5	18.20	22.79	2.06	6.15	12.66
		10	121.70	18.62	2.36	4.25	10.03
5	09 10	0	93.50	30.60	2.88	1.64	4.73
		5	12.60	25.80	1.86	3.73	6.93
		10	12.40	24.87	1.94	1.55	3.00
6	09 10	0	63.30	20.38	6.71	0.86	5.76
		5	8.90	24.87	2.27	2.01	4.54
		10	27.00	28.29	1.99	2.40	4.78
7	09 10	0	3.40	31.58	2.27	1.63	3.70
		5	31.20	32.51	2.67	3.35	8.95
		10	4.40	26.81	2.71	2.63	7.11
8	09 10	0	3.90	26.80	2.53	2.20	5.57
		5	213.30	25.64	2.19	3.83	8.38
		10	103.90	23.48	2.01	0.80	1.62

Table 5. Comparison of uptake parameters before and after the surface release of oil. Water samples were collected at stations 1 and 2 in bay 11 on 18 and 21 August 1981.

Date	Stn. no.	Depth	V_{max}	T	(K+S)
		m	$\mu g\ L^{-1}\ d^{-1}$	d	$\mu g\ L^{-1}$
08 18	1	0	4.05	2.28	9.21
		5	3.83	1.72	6.58
		10	2.74	1.77	4.86
	2	0	4.15	1.29	5.34
		5	4.13	0.89	3.67
		10	2.15	2.28	4.90
08 21	1	0	2.03	4.43	9.00
		5	1.97	4.22	8.30
		10	3.04	7.13	21.69
	2	0	3.04	2.35	7.15
		5	3.43	4.08	14.00
		10	2.35	7.89	18.54

Table 6. Hydrocarbon determinations from water samples collected at Cape Hatt on 27 and 28 August 1981. Samples were collected at various points in bays 9 and 10 during and after the dispersed oil release. Sample analyses were performed and data supplied by Seakem Oceanography Ltd.

Date	Sampling Location *	Depth	Field Fluorimetry	Fluorimetry Calibration
		m	mg L^{-1}	mg L^{-1}
08 27	6	4	2.10	0.53
	5	10	0.70	--
	Discharge Pipe	10	-- †	193, 238
	4	10	0.80	0.64
	Centre bay 9	4	17.00	--
08 28	5	11	0.45	--
	5	10	1.10	--
	6	9	0.41	0.69
	4	10	0.40	--
	4	8	0.95	0.77, 1.16
	3	7	0.50	--

* Numbers refer to microbiology station numbers

† Not reliable (saturated)

Table 7. Determinations of maximum velocity (V_{max}), turnover (T) and (K+S)(see text) of glutamic acid uptake from water samples collected at Cape Hatt on 27 and 28 August 1981. Samples were collected at various points in bays 9 and 10 during and after the dispersed oil release. Subsamples were collected for hydrocarbon determinations (see Table 6).

Date	Sampling Location *	Depth m	V_{max} $\mu g\ L^{-1}\ d^{-1}$	T d	(K+S) $\mu g\ L^{-1}$
08 27	6	5	3.13	1.30	4.08
	6	4	14.45	0.46	6.60
	5	10	4.45	1.44	6.42
	Discharge Pipe	10	1.38	9.60	13.25
	4	10	3.05	2.50	7.62
	Centre bay 9	4	1.09	8.54	9.31
08 28	5	11	3.82	1.82	6.95
	5	10	2.19	2.49	5.47
	6	9	1.92	4.62	8.90
	4	10	4.49	2.04	9.17
	4	8	3.51	2.12	7.43
	3	7	4.73	1.93	9.15

* Numbers refer to microbiology station numbers

† Control

Table 8. Mineralization of glutamic acid in the presence of 0.1% by volume weathered Norman Wells (N.W.) petroleum crude with and without Corexit 9527 supplementation. Samples from a depth of 5 m at stations 3 and 7 were collected at Cape Hatt during August and September 1981. Subsamples were processed without supplementation to serve as controls.

	V_{max}*	T*	(K+S)*
	$\mu g\ L^{-1}\ d^{-1}$	d	$\mu g\ L^{-1}$
Control	1.26 ± 0.20	4.89 ± 0.81	5.75 ± 0.81
N.W.	0.79 ± 0.24	14.91 ± 7.33	8.53 ± 3.01
Corexit	0.52 ± 0.23	48.75 ± 14.65	13.51 ± 3.13
N.W. + Corexit	0.35 ± 0.07	73.88 ± 15.76	21.56 ± 2.90

* Means of 5 samples

Table 9. Most probable number (MPN) determinations of oleoclastic cells and maximum disintegrations per minute (dpm) obtained from water samples incubated with ^{14}C-hexadecane. Samples were collected from 5 m at Cape Hatt during August and September 1981 and replicates were incubated for 20, 40 and 60 days with ^{14}C-hexadecane, 22% weathered Lago Medio (L.M.) petroleum crude and inorganic nutrients. Similar samples supplemented with Corexit 9527 were also incubated for 60 days. Results were corrected for aliquot volume and expressed per litre of sample volume.

	Stn. no.	Date	20 days dpm L^{-1} (10^{-4})	40 days dpm L^{-1} (10^{-4})	60 days dpm L^{-1} (10^{-4})	60 days (Corexit 9527) dpm L^{-1} (10^{-4})	Oleoclasts no. L^{-1} (10^{-3})
Bay 7	7	08 05	0.21	0.38	0.67	2.04	1.075
	8		0	0.20	0.24	0.86	0.525
	7	08 10	0	22.82	8.94	1.51	≥60.000
	8		0	30.55	10.27	1.12	≥60.000
	7	08 21	0	0.14	1.21	1.82	1.075
	8		0	0.47	10.88	0	6.000
	7	08 27	0	0.99	11.99	1.93	≥60.000
	8		0	0.32	4.68	1.27	5.250
	7	09 03	0	0.37	5.25	1.16	6.000
	8		0	1.48	31.25	1.47	≥60.000
	7	09 10	91.87	0.28	1.58	0.87	≥60.000
	8		268.50	0.94	17.66	1.40	≥60.000

Table 9 (cont'd)

	Stn. no.	Date	20 days dpm L^{-1} (10^{-4})	40 days dpm L^{-1} (10^{-4})	60 days dpm L^{-1} (10^{-4})	60 days (Corexit 9527) dpm L^{-1} (10^{-4})	Oleoclasts no. L^{-1} (10^{-3})
Bay 9	5	08 05	0.21	0.15	3.45	1.16	0.525
	6		0	11.60	46.50	9.56	1.075
	5	08 10	0	135.24	6.35	0.67	≥60.000
	6		0	98.78	14.17	4.34	≥60.000
	5	08 21	0	0	11.64	1.18	0.175
	6		0.61	6.79	68.65	4.74	11.500
	5	08 29	0	2.48	15.88	1.64	2.325
	6		0	0.79	15.44	1.23	0.375
	5	09 03	0	0.14	48.90	0.80	11.500
	6		0	0.47	61.76	1.42	≥60.000
	5	09 10	265.45	0.41	15.03	3.38	≥60.000
	6		149.68	0.57	10.34	1.22	≥60.000

Table 9 (cont'd)

	Stn. no.	Date	20 days dpm L^{-1} (10^{-4})	40 days dpm L^{-1} (10^{-4})	60 days dpm L^{-1} (10^{-4})	60 days (Corexit 9527) dpm L^{-1} (10^{-4})	Oleoclasts no. L^{-1} (10^{-3})
Bay 10	3	08 03	0.02	0.29	55.70	2.79	5.250
	4		0.43	0.19	18.83	1.53	2.325
	3	08 12	0	24.85	102.04	1.05	≥60.000
	4		0	7.01	63.64	0.76	≥60.000
	3	08 19	3.46	17.12	34.64	6.78	≥60.000
	4		3.34	3.89	20.85	10.76	1.875
	3	08 29	0	0.44	9.98	3.90	6.000
	4		0	0.07	5.94	1.47	2.325
	3	09 05	0.08	2.45	74.04	1.62	27.500
	4		0	1.41	6.25	1.56	≥60.000
	3	09 12	0.26	0.99	10.02	1.89	≥60.000
	4		0	6.08	30.64	0.89	≥60.000

Table 9 (cont'd)

	Stn. no.	Date	20 days dpm L^{-1} (10^{-4})	40 days dpm L^{-1} (10^{-4})	60 days dpm L^{-1} (10^{-4})	60 days (Corexit 9527) dpm L^{-1} (10^{-4})	Oleoclasts no. L^{-1} (10^{-3})
Bay 11	1	08 03	1.32	7.66	97.42	26.85	0.525
	2		0.97	2.00	151.13	3.59	27.500
	1	08 12	0	4.33	86.46	1.00	≥60.000
	2		0	1.56	95.71	0.85	≥60.000
	1	08 18	0.02	3.96	37.56	1.06	11.500
	2		0.42	0.77	3.87	1.48	11.500
	1	08 27	0	0.45	1.93	2.10	≥60.000
	2		0	0	3.32	0.89	11.500
	1	09 05	0	1.19	5.01	1.57	6.000
	2		0	1.40	13.85	0.66	≥60.000
	1	09 12	0.01	1.90	97.16	0.66	≥60.000
	2		0	0.40	8.48	11.05	≥60.000

Table 10. Comparison of disintegrations per minute (dpm) obtained from water and sediment suspension samples incubated with ^{14}C-hexadecane in the presence of different crudes. Samples were incubated for 20, 40 and 60 days with radiolabelled hexadecane, weathered Norman Wells (N.W.), 22% weathered Lago Medio (L.M.) and Norwegian weathered Lago Medio (N.L.M.) (see text) petroleum crudes and inorganic nutrients. Data are expressed in dpm L^{-1} (10^{-4}) and dpm g^{-1} dry weight (10^{-3}) for water and sediment suspension samples respectively.

Water

Date	Hexadecane N.W.			Hexadecane L.M.			Hexadecane N.L.M.		
	20 days	40 days	60 days	20 days	40 days	60 days	20 days	40 days	60 days
08 05	0.0	1.965	26.143	0.0	11.599	46.495	--	--	--
08 10	0.0	0.791	4.372	0.0	98.784	14.173	0.0	6.671	266.119
08 21	0.0	0.278	0.719	0.610	67.908	68.654	--	--	--
08 29	0.0	0.005	0.088	0.0	0.790	15.441	0.0	1.789	45.355
09 03	0.0	0.653	12.121	0.0	0.474	61.756	0.0	2.426	42.110
09 10	75.349	2.296	14.041	149.681	0.569	10.345	--	--	--

Sediment suspension

Date	Hexadecane N.W.			Hexadecane L.M.			Hexadecane N.L.M.		
	20 days	40 days	60 days	20 days	40 days	60 days	20 days	40 days	60 days
08 09	0.088	0.364	0.455	0.440	0.614	57.545	--	--	--
08 18	0.295	0.482	0.953	0.442	1.600	106.134	--	--	--
08 25	0.423	5.857	0.0	0.085	0.294	28.359	0.055	0.712	74.371
08 31	0.165	11.244	18.369	0.161	0.968	36.234	0.089	0.628	25.513
09 06	0.145	28.344	27.177	0.104	3.977	68.141	--	--	--
09 13	0.054	0.515	87.545	0.115	0.352	1.915	--	--	--
09 19	0.105	0.588	34.850	0.085	5.271	14.738	--	--	--

Table 11. Comparison of disintegrations per minute (dpm) obtained from water and sediment suspension samples incubated with ^{14}C-hexadecane or ^{14}C-phenanthrene. Samples were incubated for 20, 40 and 60 days with radiolabelled hexadecane or phenanthrene, weathered Lago Medio petroleum crude and inorganic nutrients. Data are expressed in dpm L^{-1} (10^{-4}) and dpm g^{-1}dry weight (10^{-3}) for water and sediment suspension samples respectively.

Water

Date	Hexadecane			Phenanthrene		
	20 days	40 days	60 days	20 days	40 days	60 days
08 05	0.0	11.599	46.495	0.0	0.0	0.259
08 10	0.0	98.784	14.173	0.0	16.639	34.052
08 21	0.610	6.791	68.654	0.0	0.281	11.132
08 29	0.0	0.790	15.441	0.0	0.419	52.835
09 03	0.0	0.474	61.756	0.0	0.0	3.878
09 10	149.681	0.569	10.345	0.0	0.0	2.663

Sediment suspension

Date	Hexadecane			Phenanthrene		
	20 days	40 days	60 days	20 days	40 days	60 days
08 09	0.440	0.614	57.545	0.144	0.048	0.036
08 18	0.442	1.600	106.134	0.026	0.111	0.179
08 25	0.085	0.294	28.359	0.0	0.013	0.664
08 31	0.161	0.968	36.234	0.0	0.0	0.0
09 06	0.104	3.977	68.141	0.0	0.0	0.0
09 13	0.115	0.352	1.915	0.0	0.0	0.0
09 19	0.085	5.271	14.738	0.0	0.0	0.0

Table 12. Determinations of colony-forming units (CFU), total counts (TC) of bacterial cells, maximum velocity (V_{max}), turnover (T) and (K+S) (see text) of glutamic acid uptake from sediment suspension samples collected from surface sediment at Cape Hatt during August and September 1981. Quantities are expressed per gram dry weight of sediment.

Station	Date	CFU	TC	V_{max}	T	(K+S)
		no. g^{-1} (10^{-5})	no. g^{-1} (10^{-7})	$\mu g\ g^{-1}\ d^{-1}$	d	$\mu g\ g^{-1}$
1	08 10	3.10	95.23	6.71	2.18	14.60
2	08 10	4.40	48.65	7.31	2.66	19.44
3	08 07	8.60	63.76	10.23	3.82	27.49
4	08 07	15.20	77.88	10.01	1.50	15.03
5	08 09	7.20	49.89	5.45	3.15	17.17
6	08 09	4.80	70.69	10.34	2.46	25.46
7	08 07	16.30	72.25	14.90	1.59	23.68
8	08 07	65.00	140.70	15.93	1.32	21.02
1	08 14	13.60	149.55	58.95	0.82	27.69
2	08 14	9.70	69.83	40.82	0.88	35.96
3	08 14	6.00	99.77	13.58	1.61	21.59
4	08 14	11.40	171.42	21.22	1.45	30.75
5	08 18	10.40	151.29	14.67	1.58	23.16
6	08 18	8.80	75.18	17.81	1.76	31.31
7	08 16	190.90	135.47	63.86	0.51	32.69
8	08 16	177.20	116.10	45.58	0.52	23.85

Table 12 (cont'd)

Station	Date	CFU	TC	V_{max}	T	(K+S)
		no. g^{-1} (10^{-5})	no. g^{-1}(10^{-7})	µg g^{-1} d^{-1}	d	µg g^{-1}
1	08 23	15.60	120.79	21.60	0.96	20.79
2	08 23	19.70	109.61	25.33	1.15	28.91
3	08 23	16.90	109.79	12.86	1.37	17.91
4	08 23	14.70	85.46	12.72	2.03	25.84
5	08 24	15.20	54.20	9.73	2.36	22.87
6	08 24	14.80	78.71	ND	ND	ND
7	08 23	80.40	109.08	49.71	0.78	38.71
8	08 23	52.90	113.43	27.69	0.79	21.81
7	08 26	89.60	110.37	50.29	0.98	46.91
8	08 26	27.60	340.21	17.63	0.89	15.70
1	08 31	11.80	172.78	38.66	1.00	38.52
2	08 31	14.40	137.06	36.63	0.81	29.84
3	08 31	9.20	97.83	16.80	1.35	22.48
4	08 31	17.20	171.76	20.27	1.04	21.20
5	08 31	35.80	103.65	13.81	1.34	18.57
6	08 31	28.60	61.51	9.79	1.51	14.81
7	09 01	35.00	110.82	16.99	1.32	21.56
8	09 01	50.00	191.04	19.21	1.90	36.37

Table 12 (cont'd)

Station	Date	CFU no. g^{-1} (10^{-5})	TC no. g^{-1} (10^{-7})	V_{max} $\mu g\ g^{-1}\ d^{-1}$	T d	(K+S) $\mu g\ g^{-1}$
1	09 06	8.00	93.27	24.64	1.05	25.94
2	09 06	12.00	104.21	53.05	0.40	21.49
3	09 06	15.10	54.63	16.93	0.92	15.64
4	09 06	16.90	98.08	20.24	0.80	16.04
5	09 06	30.10	68.64	16.86	1.01	16.93
6	09 06	12.40	37.54	ND	ND	ND
7	09 06	59.70	135.62	44.96	0.70	28.10
8	09 06	58.40	232.45	59.78	0.68	41.04
1	09 13	4.50	61.42	12.11	1.70	20.75
2	09 13	8.50	88.36	15.60	1.61	25.13
3	09 13	8.50	63.29	8.84	2.11	18.62
4	09 13	9.70	59.20	10.24	1.61	16.50
5	09 13	12.50	70.04	10.84	1.15	12.46
6	09 13	10.60	51.87	14.29	1.71	24.33
7	09 13	27.00	93.04	19.28	1.24	21.19
8	09 13	26.30	199.89	49.53	0.74	36.87

Table 12 (cont'd)

Station	Date	CFU	TC	V_{max}	T	(K+S)
		no. g^{-1} (10^{-5})	no. g^{-1} (10^{-7})	$\mu g\ g^{-1}\ d^{-1}$	d	$\mu g\ g^{-1}$
1	09 19	9.30	101.48	14.78	1.46	21.65
2	09 19	7.60	97.18	19.45	1.59	30.83
3	09 19	12.20	108.07	11.06	1.62	17.84
4	09 19	11.60	106.08	12.37	1.79	22.13
5	09 19	16.30	106.00	11.12	1.93	21.42
6	09 19	16.40	80.44	14.91	2.12	31.62
7	09 19	60.60	168.13	50.07	0.99	46.53
8	09 19	126.80	189.87	58.03	0.78	45.23

Table 13. Most probable number (MPN) determinations of oleoclastic cells and maximum disintegrations per minute (dpm) obtained from sediment suspension samples incubated with ^{14}C-hexadecane. Samples were collected from surface sediment at Cape Hatt during August and September 1981 and replicates were incubated for 20, 40 and 60 days with ^{14}C-hexadecane, 22% weathered Lago Medio (L.M.) petroleum crude and inorganic nutrients. Similar samples supplemented with Corexit 9527 were also incubated for 60 days. Results were corrected for dilution and expressed per gram dry weight of sediment.

	Stn. no.	Date	20 days dpm g^{-1} (10^{-3})	40 days dpm g^{-1} (10^{-3})	60 days dpm g^{-1} (10^{-3})	60 days (Corexit 9527) dpm g^{-1} (10^{-3})	Oleoclasts no. g^{-1} (10^{-3})
Bay 7	7	08 07	0.011	0.813	21.298	1.076	≥3.354
	8		0.020	0.835	1.455	1.779	≥4.939
	7	08 16	0.171	0.404	0.207	0.282	≥4.473
	8		0.211	4.686	113.284	1.093	≥4.264
	7	08 23	0.506	5.308	18.333	1.600	≥7.204
	8		0.223	1.582	47.131	2.846	0.557
	7	08 26	0.246	0.592	4.103	1.041	2.593
	8		0.150	0.620	8.334	0.777	≥5.585
	7	09 01	0.085	0.320	4.835	0.866	0.245
	8		0.280	0.557	15.383	0.512	0.737
	7	09 06	0.118	0.302	0.828	0.653	≥3.264
	8		0.283	1.222	7.033	4.117	≥5.361

Table 13 (cont'd)

	Stn. no.	Date	20 days dpm g^{-1} (10^{-3})	40 days dpm g^{-1} (10^{-3})	60 days dpm g^{-1} (10^{-3})	60 days (Corexit 9527) dpm g^{-1} (10^{-3})	Oleoclasts no. g^{-1} (10^{-3})
Bay 7	7	09 13	0.078	0.426	18.544	0.351	0.622
	8		0.107	2.535	53.952	9.050	0.054
	7	09 19	0.054	0.565	49.591	1.290	0.418
	8		0.043	1.258	68.736	2.798	$\geq$5.662
Bay 9	5	08 09	0.179	0.624	26.790	1.513	$\geq$3.483
	6		0.440	0.614	57.545	0.460	1.634
	5	08 18	0.395	0.607	4.426	0.740	$\geq$3.466
	6		0.442	1.600	106.134	0.792	$\geq$3.776
	5	08 24	0	2.290	57.167	0.349	0.135
	6		0.085	0.294	28.359	0.449	0.060
	5	08 31	0.126	0.589	4.588	0.465	1.581
	6		0.161	0.968	36.234	0.602	1.733
	5	09 06	0.141	0.489	20.331	0.790	$\geq$3.683
	6		0.104	3.977	68.141	0.755	0.362

Table 13 (cont'd)

	Stn. no.	Date	20 days dpm g^{-1} (10^{-3})	40 days dpm g^{-1} (10^{-3})	60 days dpm g^{-1} (10^{-3})	60 days (Corexit 9527) dpm g^{-1} (10^{-3})	Oleoclasts no. g^{-1} (10^{-3})
Bay 9	5	09 13	0	1.318	5.790	0.482	0.288
	6		0.115	0.352	1.915	0.489	0.337
	5	09 19	0.070	0.408	60.447	0.813	0.204
	6		0.085	5.271	14.738	1.805	0.684
Bay 10	3	08 07	0.005	0.591	0.950	0.945	$\geq$3.568
	4		0	0	0	0.195	.001
	3	08 14	0.020	0.349	2.259	0.404	$\geq$3.682
	4		0.065	0.720	8.446	0.726	$\geq$4.511
	3	08 23	0.296	9.465	8.919	1.248	$\geq$3.995
	4		0.275	8.518	26.945	0.856	0.391
	3	08 31	0	0.744	2.100	0.527	0.683
	4		0.096	0.612	1.759	0.525	$\geq$3.467
	3	09 06	0.061	0.407	1.281	0.363	0.064
	4		0.185	6.196	1.684	0.561	0.112

Table 13 (cont'd)

	Stn. no.	Date	20 days dpm g^{-1} (10^{-3})	40 days dpm g^{-1} (10^{-3})	60 days dpm g^{-1} (10^{-3})	60 days (Corexit 9527) dpm g^{-1} (10^{-3})	Oleoclasts no. g^{-1} (10^{-3})
Bay 10	3	09 13	0.094	0.528	35.158	0.377	≥3.412
	4		0.051	0.402	35.059	0.456	0.339
	3	09 19	0.085	0.384	42.586	1.877	1.617
	4		0.109	0.564	59.317	0.659	0.139
Bay 11	1	08 10	0.029	1.165	0.855	0.957	≥3.080
	2		0.073	0.678	0.582	0.790	≥3.110
	1	08 14	0.134	1.635	41.582	1.705	≥7.920
	2		0.035	1.306	201.165	0	≥5.322
	1	08 23	0.173	3.312	89.157	1.162	0.177
	2		0.196	3.820	91.826	9.168	≥4.549
	1	08 31	0.240	0.535	4.148	1.628	1.644
	2		0	0.436	6.931	0.736	0.146
	1	09 06	0.098	6.088	34.396	0.769	≥3.313
	2		0.112	0.999	175.691	0.657	0.085

Table 13 (cont'd)

	Stn. no.	Date	20 days dpm g^{-1} (10^{-3})	40 days dpm g^{-1} (10^{-3})	60 days dpm g^{-1} (10^{-3})	60 days (Corexit 9527) dpm g^{-1} (10^{-3})	Oleoclasts no. g^{-1} (10^{-3})
Bay 11	1	09 13	0.126	0.306	22.630	0.762	0.057
	2		0.096	0.450	20.085	0.467	1.471
	1	09 19	0.132	0.324	13.803	1.617	0.671
	2		0.080	0.554	52.604	6.001	0.318